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Archives of Internal Medicine

VOLUME 40 AUGUST, 1927 NUMBER 2

CALCIUM STUDIES IN JAUNDICE

WITH SPECIAL REFERENCE TO THE EFFECT OF PARATHYROID EXTRACT ON THE DISTRIBUTION OF CALCIUM *

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AND

BURGESS GORDON, M.D.

PHILADELPHIA

In recent years considerable progress has been made in the study of jaundice, but comparatively little is known about the details of the chemical processes involved. The tendency to prolonged bleeding in jaundiced patients has long been known. Obviously this fact is of practical importance, since it adds greatly to the operative risk in a condition that often requires surgical treatment. It is generally believed that the hemorrhagic tendency is intimately related to some fault in the availability and utilization of calcium, although the exact mechanism is far from being well understood.

REVIEW OF PREVIOUS OBSERVATIONS

Calcium exists in the blood stream in several forms: ionized and unionized, diffusible and nondiffusible, free and combined. According to Howland,[1] calcium, phosphoric acid and carbon dioxide are present in the blood in a finely balanced equilibrium, illustrated by the following equation:

$$Ca_3(PO_4)_2 + 2H_2CO_3 \rightleftarrows 2CaHPO_4 + Ca(HCO_3)_2 \rightleftarrows 2Ca^{++} + 2HPO_4^{--} + Ca^{++} + 2HCO_3^-$$

Wells [2] states that the calcium salts are held partly in solution, partly in suspension and partly in the form of calcium ion protein compounds. This condition is extremely unstable; it is subject to alteration by slight changes in any of the several controlling factors. In addition, it is now recognized that the parathyroid hormone plays an important part in the maintenance of the normal calcium level and distribution.

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* From the Medical Service of Dr. Thomas McCrae and the Department for Diseases of the Chest, Jefferson Hospital.

* A part of the expense for this investigation was defrayed by a grant from Mr. L. A. Swayze, of Canton, Pa.

* Read before the College of Physicians of Philadelphia, June 1, 1927.

1. Howland, J.: Etiology and Pathogenesis of Rickets, Harvey Lectures **18**:189, 1922-1923.

2. Wells, H. G.: Chemical Pathology, Philadelphia, W. B. Saunders Company, 1925, p. 492.

Kirk and King [3] have shown that normally all the common inorganic constituents of serum are diffusible except calcium, of which only from 70 to 75 per cent is diffusible. Grove and Vines [4] state that calcium is present in ionized and combined forms, but that under normal conditions only the ionized form is found in the serum after coagulation, which suggests the importance of combined calcium in the mechanism of coagulation. With such a variety of factors operating to maintain the normal relations of calcium in the blood stream, it is evident that a disturbing element may exercise its effect in several ways. Anything that will alter the calcium level, the hydrogen ion concentration, the phosphate and the carbon dioxide content, the amount and nature of the proteins, the ratio of diffusible to nondiffusible calcium and of the ionized to the combined forms will cause some disturbance in the functional availability and utilization of calcium. This, however, is not necessarily manifested by any change in the phenomenon of coagulation.

The characteristic feature of jaundice is the presence of an excess of bile pigments and bile salts in the blood. Interference with the normal mechanism of coagulation is perhaps due to the action of these substances, although in advanced cases, hepatic malfunction may play a part through the associated diminution in the formation of fibrinogen. Prolongation of the coagulation time does not occur in all patients with jaundice. Lee and Vincent [5] state that it is met with only to a slight extent in catarrhal jaundice. These authors have observed no change in coagulation time until the jaundice was of five weeks' duration, and they conclude that such change depends on the intensity of the condition and the amount of available tissue calcium. However, as is well known, the hemorrhagic tendency in jaundice is not incompatible with a normal clotting and bleeding time.

Haessler and Stebbins [6] believe that in some way the bile salts retard the conversion of fibrinogen to fibrin. This factor possibly plays a minor part, however, and for our purposes may be disregarded. Interest is particularly focused on the alteration in the distribution and functional availability of calcium caused by the presence of abnormal amounts of bile pigments in the blood and tissues.

The exact manner in which the pigments operate to diminish the available circulating calcium is unsettled. King and Stewart [7] report

3. Kirk, P. L., and King, C. G.: Calcium Distribution in the Blood, J. Lab. & Clin. Med. **11**:928 (July) 1926.

4. Grove, W. R., and Vines, H. W. C.: Calcium Deficiency and Treatment by Parathyroid Extract, Brit. M. J. **1**:791 (May 20) 1922.

5. Lee, R. I., and Vincent, B.: The Relation of Calcium to the Delayed Coagulation of Blood in Jaundice, Arch. Int. Med. **16**:59 (July) 1915.

6. Haessler, H., and Stebbins, M. G.: Effect of Bile on Clotting Time of Blood, J. Exper. Med. **29**:445, 1919.

7. King, J. H., and Stewart, H. A.: Effect of the Injection of Bile on the Circulation, J. Exper. Med. **11**:673, 1909.

that the blood calcium is increased in jaundice owing to obstruction. This observation was confirmed by King, Bigelow and Pearce.[8] They believe that calcium combines with the pigments as a protective measure in an attempt to counteract their toxicity. This combination appears to fix the calcium in some way and to render it functionally unavailable, probably by lessening the degree of diffusibility and ionization. Kirk and King[3] have found that whereas normally 72.3 per cent of the blood calcium is diffusible, in jaundice this fraction is reduced to 55.2 per cent. They state that the percentage of diffusible calcium in jaundice is subnormal, roughly, in proportion to the degree of jaundice. They add that this is not the only factor that increases the coagulation time, as similar changes in diffusibility occur in other conditions without affecting coagulation.

It is not unlikely that the nature and amount of blood proteins are of importance in this connection. In certain forms of hemolytic jaundice and in some cases of pernicious anemia the bilirubin content of the blood may be far above the normal renal threshold concentration without appearing in the urine. Bowler and Walters[9] believe that the pigment in such cases is fixed to the plasma proteins to some extent, lessening the diffusibility of the pigment molecule and preventing its excretion. The protein-pigment combination, uniting with calcium, increases the ratio of protein bound to free calcium, altering the normal diffusibility and ionic relations.

When pigment deposition occurs, a considerable amount of calcium which is normally circulating and utilized is fixed in the tissues. Early in the condition this fixation is counterbalanced by the normal calcium reserves, but as the intensity progresses and the degree of fixation increases, these reserves become relatively depleted, and in a functional sense, an actual calcium deficiency occurs. This accounts for the fact, observed by Lee and Vincent,[5] that early in jaundice there is no change in coagulation time. As they state, such alteration is largely dependent on the amount of calcium in the tissues that can be taken into the blood.

In an attempt to supply this deficiency in jaundice, calcium therapy has been employed with variable results. The success of oral administration is dependent on so many factors that uniformly good results cannot be expected. Lee and Vincent[5] report that large doses of calcium lactate must be given over a period of several days before any alteration in clotting time occurs. Bowler and Walters[10] use small amounts of calcium chloride intravenously for a short period before

8. King, J. H.; Bigelow, J. E., and Pearce, L.: Experimental Obstructive Jaundice, J. Exper. Med. **14**:159, 1911.

9. Bowler, J. P., and Walters, W.: Toxicity and Rate of Excretion of Calcium Chloride from the Blood Stream, Ann. Surg. **80**:545, 1924.

10. Walters, W., and Bowler, J. P.: Preoperative Preparation of Patients with Obstructive Jaundice, Surg. Gynec. Obst. **39**:201, 1924.

operation, with apparent success. They report the interesting observation that twice the amount of calcium chloride injected intravenously is required to raise the blood serum calcium in jaundice to the same level as in normal persons. This, as they say, suggests a calcium deficiency that is not otherwise apparent.

Parathyroid extracts have been proved to exert a profound influence on calcium metabolism. Their administration results in an increase in the amount of available circulating calcium and also perhaps aids in its utilization. In a previous study [11] we observed the changes in the calcium content of the whole blood and of serum at three hour intervals following the injection of parathyroid extract. The results in six normal cases are shown in table 1. It was noted that the rise in serum calcium generally preceded that in whole blood. Furthermore, it was observed that the curve of whole blood calcium differs materially from that of serum, but corresponds more exactly than the latter to the curve

TABLE 1.—*Coincident Determinations of the Whole Blood and Serum Calcium in Nonjaundiced Patients at Three Hour Intervals Following the Administration of Parathyroid Extract*

	Control		3 Hours		6 Hours		9 Hours		12 Hours	
Case	Blood*	Serum*	Blood	Serum	Blood	Serum	Blood	Serum	Blood	Serum
1.....	7.64	10.4	7.7	15.7	9.8	11.4	10.05	11.4	12.24	10.9
2.....	6.43	10.45	11.36	10.22	10	9.04	10.28	10.4	8.62	10.24
3.....	6.6	10.2	6.2	11.3	8.1	11.3	8.1	10.8	11.1	9.6
4.....	6.52	10.43	5.7	14.54	9.52	11.4	11.2	11.04	10.34	10.76
5.....	9.54	10.45	11.99	10	12.2	9.5	9.8	10.78	8.07	9.13
6.....	9.61	10.09	10.09	10.09	10.52	8.8	11.17	9.52	8.18	9.04

* Calcium values are expressed in milligrams per hundred cubic centimeters.

of changes in the coagulation time. From this we concluded that the whole blood or plasma calcium content was a more accurate index of changes in the blood following the administration of parathyroid hormone than the serum calcium.

METHODS

The method used for the determination of whole blood calcium [12] was based on the Clark-Collip modification of the Kramer-Tisdall method for determining serum calcium.[13] The figures are undoubtedly higher than the actual calcium content, owing to the reaction of certain constituents of the stroma of the corpuscles. However, the method was

11. Cantarow, A.; Caven, W. R., and Gordon, B.: Changes in the Chemical and Physical Characteristics of the Blood Following the Administration of Parathyroid Hormone, Arch. Int. Med. **38**:502 (Oct.) 1926.

12. Caven, W. R., and Cantarow, A.: A Method for the Determination of Calcium in Whole Blood, J. Lab. & Clin. Med. **12**:76 (Oct.) 1926.

13. Clark, E. P., and Collip, J. B.: A Study of the Tisdall Method for the Determination of Blood Serum Calcium with a Suggested Modification, J. Biol. Chem. **63**:461 (March) 1925.

used to obtain a curve showing the changes in blood calcium after the injection of parathyroid extract rather than to determine the absolute calcium values. It was found satisfactory for this purpose, since the error was a constant one. This was proved subsequently by simultaneous determinations of plasma calcium, coagulation being retarded by the use of heparin.[14] The curve showing the changes in the plasma calcium was found almost to parallel that of the whole blood.

The present investigation was undertaken to compare the response to parathyroid extract [15] in jaundiced and in normal persons. The series consists of fourteen cases of jaundice; one was hemolytic in type, three were cases of hepatitis due to arsphenamine and ten were of an obstructive nature, resulting from either gallstones or carcinoma of the head of the pancreas. The intensity of the jaundice varied from 2 to 20 units

TABLE 2.—*Coincident Determinations of the Whole Blood and Serum Calcium in Jaundice at Three Hour Intervals Following the Administration of Parathyroid Extract*

			Control		3 Hours		6 Hours		9 Hours		12 Hours	
Case	Type	Units	Blood*	Serum*	Blood	Serum	Blood	Serum	Blood	Serum	Blood	Serum
1	Hemolytic†	2	10.45	10.45	12.72	11.36	11.17	9.6	9.16	9.31	10.26	8.7
2	Obstructive	16	10.59	10	9.54	9.54	10.18	9.8	10.53	8.43	12.5	9.13
3	Obstructive	10	12	10.02	11.8	10.8	11.2	14.1	11.6	12	.....	....
4	Obstructive	3	11	9.52	10.5	7.6	13	10	12	10	.....	....
5	Obstructive	15	4.8	10	7.9	10	10.9	10.9	11.6	10.9	.....	....
6	Obstructive	20	7.5	9.3	8.5	9.7	8.8	9.7	9.7	9.7	8.5	9.7
7	Obstructive	8	8.1	9.55	11.36	9.62	11.47	11	14.75	10.4	.....	....
8	Obstructive	7	8.2	11.3	10.2	12.1	11.5	12.4	10	11.6	.....	....
9	Obstructive	10	4.9	10.8	9.1	13.1	11.3	12.2	8.4	10	.....	....
10	Obstructive	15	8.2	9.3	.....	10	.....	10	.....	9.7	.....	....
11	Obstructive	10	10.96	10.32	.....	.....	.....	.....	.....	.....	.....	....
12	Toxic hepatitis‡	8	4.9	10.5	12	11	12.4	11	11.7	9	.....	....
13	Toxic hepatitis.	14	7.6	12	8.3	13	8.3	13.5	10	13.5	10.5	14.1
14	Toxic hepatitis.	4	8.2	9.5	.....	9.5	.....	10.5	.....	10.4	.....	....

* Calcium values are expressed in milligrams per hundred cubic centimeters.
† Amount of bilirubin expressed in units by the van den Bergh method.
‡ Caused by arsphenamine.

of bilirubin as determined by the van den Bergh test. The following determinations were made: clotting time by the Boggs method, and the calcium content of whole blood,[12] serum [13] and, in some instances, plasma. After one control observation the determinations were repeated at three hour intervals following the intramuscular injection of parathyroid extract. The figures for calcium are presented in tables 2 and 3.

RESULTS OF AUTHORS' INVESTIGATIONS

Coagulation Time.—The clotting time in all cases before administration of parathyroid hormone was well within normal limits. It ranged between three and five minutes, with apparently no relation to the depth,

14. Plasma determinations were made by the Clark-Collip method, powdered heparin being used to prevent coagulation in the specimen of blood.

15. The preparation used was para-thor-mone, supplied by Eli Lilly and Company through the kindness of Dr. G. H. A. Clowes and Dr. J. H. Warvel.

duration or type of jaundice. The response to the administration of the hormone was similar to that previously observed in persons who were not jaundiced.[11] The clotting time was reduced in all instances, the degree of reduction varying between 5 and 65 per cent. The maximum effect occurred from six to twelve hours after the injection. The rapidity of coagulation did not bear as close a relationship to the whole blood calcium as it did in the normal cases, but it still appeared to be much more dependent on this factor than on the serum calcium level.

Serum Calcium.—The normal range of serum calcium is from 9 to 11 mg. per hundred cubic centimeters. The control values in the normal cases were from 10.09 to 10.45 mg. After the administration of parathyroid extract, there was a tendency toward a rise in serum calcium within the first three hours, followed by a gradual return to normal (table 1). The maximum variation occurred three hours after the injection.

TABLE 3.—*Coincident Determinations of the Whole Blood, Plasma and Serum Calcium in Jaundice at Three Hour Intervals Following the Administration of Parathyroid Extract*

	Control			3 Hours			6 Hours			9 Hours			12 Hours		
Case	Blood*	Plasma*	Serum*	Blood	Plasma	Serum	Blood	Plasma	Serum	Blood	Plasma	Serum	Blood	Plasma	Serum
8.....	8.2	14.1	11.2	10.2	17.6	12.1	11.5	18.7	12.4	10	17.1	11.6	....	....	....
9.....	4.9	8.8	10.8	9.1	17.6	13.1	11.3	20.1	12.2	8.4	16.1	10	....	....	....
10.....	8.2	9.6	9.3	....	12	10	....	12	10	....	11.9	9.7	....	....	....
11.....	10.96	18.01	10.32	....	....	....	....	....	....	....	....	....	....	....	....
13.....	7.6	17.8	12	8.3	18.2	13	8.3	19	13.5	10	19.5	13.5	10.5	20.3	14.1
14.....	8.2	12.5	9.5	....	15	9.5	....	....	....	....	15.2	10.4	....	....	....

* Calcium values are expressed in milligrams per hundred cubic centimeters.

In the series of patients with jaundice the control values varied slightly beyond normal limits, from 9.3 to 12 mg. per hundred cubic centimeters. The response to parathyroid hormone was inconstant; in some instances no rise occurred; in others there were slight rises at varying intervals. We do not attach significance to the absence of an increase in calcium in several cases, since the rise may have occurred in an interval between our determinations. However, the results are interesting when compared with those of normal persons as illustrated in chart 1. It is seen that the limits of variation in serum calcium are slightly affected by parathyroid extract in jaundiced persons as compared with normal persons.

Whole Blood.—The whole blood calcium in the normal group varied between 6.43 and 9.61 mg. per hundred cubic centimeters.[12] After the administration of parathyroid extract, there was a gradual tendency toward an increase in the blood calcium level, the degree of variation

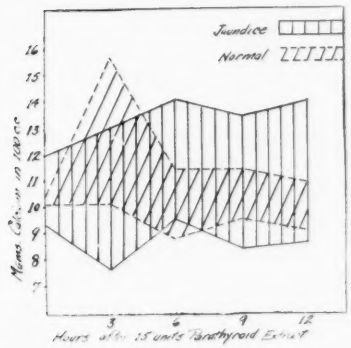

Chart 1.—Comparison between the variation in serum calcium in jaundiced and in normal persons before and at three hour intervals following the administration of parathyroid extract. The area occupied by diagonal lines represents variation between minimum and maximum serum calcium values in normal persons; the area occupied by perpendicular lines represents variation between minimum and maximum serum calcium values in jaundice.

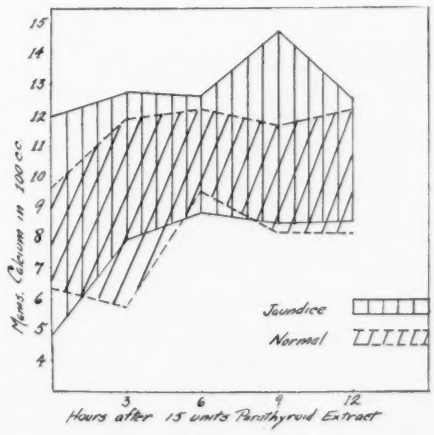

Chart 2.—Comparison between the variation in whole blood calcium in jaundiced and in normal persons before and at three hour intervals following the administration of parathyroid extract. The area occupied by diagonal lines represents variation between minimum and maximum whole blood calcium values in normal persons; the area occupied by perpendicular lines represents variation between minimum and maximum whole blood calcium values in jaundice.

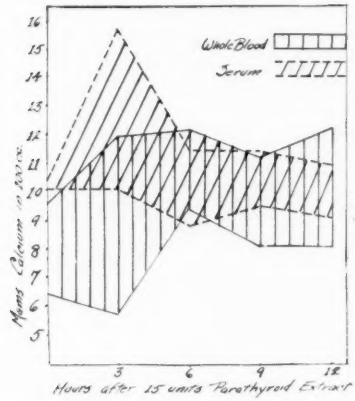

Chart 3.—Comparison between the variation in serum and whole blood calcium in normal persons before and at three hour intervals following the administration of parathyroid extract. The area occupied by diagonal lines represents variation between minimum and maximum normal serum calcium values; the area occupied by perpendicular lines represents variation between minimum and maximum normal whole blood calcium values.

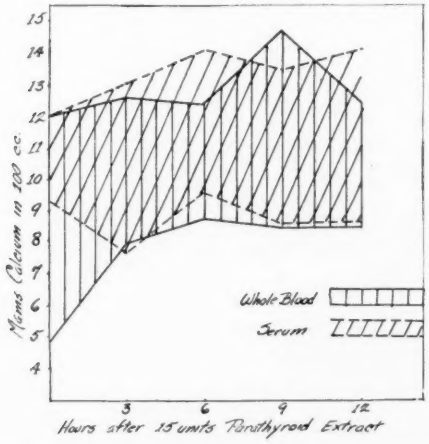

Chart 4.—Comparison between the variation in serum and whole blood calcium in jaundice before and at three hour intervals following the administration of parathyroid extract. The area occupied by diagonal lines represents variation between minimum and maximum serum calcium values in jaundice; the area occupied by perpendicular lines represents variation between minimum and maximum whole blood calcium values in jaundice.

remaining approximately the same as before. In the jaundiced patients the control values for blood calcium ranged between 4.8 and 12 mg. per hundred cubic centimeters. This wide variation was the first striking difference observed between jaundiced and normal blood. The calcium level bore no relation to the coagulation time, degree, type or duration of the jaundice. The injection of parathyroid extract was followed by a marked decrease in the degree of variation; at the end of six hours, and particularly after twelve hours, the range in jaundiced persons was practically identical with that in normal persons (chart 2). The relationship between whole blood and serum variations in normal and in jaundiced persons is illustrated in charts 3 and 4.

Plasma Calcium.—In some cases the calcium content of the plasma was determined. It varied between 8.8 and 18.01 mg. per hundred cubic centimeters in the group of patients who had jaundice. The determinations after parathyroid administration were almost exactly parallel to those of whole blood.

COMMENT

In reviewing these data several interesting facts might be noted. However, there are two points which to us seem of particular significance: first, the wide variation in the calcium content of whole blood in jaundice as compared with that of normal blood; second, the fact that after the administration of parathyroid extract this variation is altered, so that at the end of twelve hours the calcium content of the blood corresponds almost exactly to that in normal persons treated in the same manner (chart 2).

The variation in the calcium content of whole blood is undoubtedly of considerable significance. We have stated that the amount seems to be independent of the type, intensity and duration of the jaundice, and that it bears no relation to the coagulation time. Of still greater interest is the fact that it is equally independent of the serum calcium content (table 2). For example in case 2 the patient had a whole blood calcium of 10.59 mg. and in case 5, 4.80 mg. The serum calcium content in both cases was 10 mg. per hundred cubic centimeters. It seems obvious that the amount of calcium which participated actively in the process of coagulation, and which perhaps was incorporated in the clot in protein or lipiodol combination, differed greatly in the two instances, being comparatively large in case 2 and slight in case 5. This relative fixation of serum calcium irrespective of the blood calcium level is in all likelihood due to the presence of the bile pigments, which bind the calcium, rendering it functionally unavailable. Since most of the pigment remains in the serum after coagulation, the bound calcium likewise remains. This tends to maintain the relative constancy of the serum calcium values as compared with those of whole blood in jaundice. The wide

variation in the blood calcium must be dependent largely on corresponding variations in the general calcium metabolism of the tissues, in the functionally available and diffusible fractions and in the amount fixed in the tissues by the pigments and excreted with them in the urine. The supposition of functional deficiency is supported by the work of Bowler and Walters [10] referred to previously, in which they observed that much more calcium chloride, given intravenously, is necessary to raise the serum calcium in jaundice to the same level as in normal persons.

The effect of parathyroid extract on the distribution of blood calcium in jaundice is striking. Collip states [16] that in the normal animal the function of the parathyroid hormone "appears to be that of a regulator of calcium metabolism, and its action is primarily as a calcium mobilizer." We have shown that in jaundice there is a disturbance of calcium metabolism of varying degree, evidenced by the abnormal distribution of calcium in the blood stream. In twelve hours after the administration of parathyroid extract the distribution of calcium is temporarily practically identical in jaundiced and in normal persons. Evidently the hormone tends to restore the normal balance between blood and tissues in an attempt to remedy the existing functional deficiency. One evidence of the success of this attempt is the reduction of the coagulation time, suggesting an increase in the availability and utilization of the calcium of the blood. Through increasing the functionally available calcium the tendency to slow, protracted hemorrhage of a diffuse nature may be lessened, as noted in incised tissues in jaundice often associated with a normal coagulation and bleeding time. As previously suggested, this favorable effect is perhaps due [17] to a decrease in the permeability of the capillary walls, aided, undoubtedly, by the increased coagulability of the blood following administration of parathyroid extract.

CONCLUSIONS

1. In jaundice it is evident that a functional deficiency in calcium exists, probably due to the increased amount of bile pigments in the blood and tissues.

2. The calcium content of whole blood, plasma and serum and the coagulation time have been determined at three hour intervals after the injection of parathyroid extract. The observations in jaundice are compared with those in nonjaundiced persons.

3. The serum calcium values varied slightly, ranging from 9.3 to 12 mg. per hundred cubic centimeters in jaundice. There was an

16. Collip, J. B.: The Calcium Mobilizing Hormone of the Parathyroid Glands, J. A. M. A. **88**:566 (Feb. 19) 1927.

17. Gordon, B., and Cantarow, A.: The Use of Parathyroid Extract in Hemorrhage, J. A. M. A. **88**:1301 (April 23) 1927.

extremely wide variation, however, in the calcium content of whole blood, the figures ranging between 4.8 and 12 mg. per hundred cubic centimeters.

4. Twelve hours after the administration of the parathyroid hormone the variation in the whole blood calcium of jaundiced and nonjaundiced patients was practically identical.

5. Parathyroid extract acts as a mobilizer of calcium; in jaundice it tends to restore the normal distribution and functional availability of this element.

6. The favorable effect of the hormone on the tendency of jaundiced tissue to bleed is due largely to the increased coagulability of the blood and probably to the diminished permeability of the capillary walls, the result of an increase in functioning calcium.

THE STORAGE OF WATER BY VARIOUS TISSUES OF THE BODY *

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LITTLE ROCK, ARK.

While it is well known that water plays an extremely important rôle in the economy of the body, and that it is the most abundant constituent, making up 90 pounds (40.8 Kg.) of a man weighing 143 pounds (64.9 Kg.), there have been few investigations concerning the exact distribution of this water in the animal as a whole, although the water content of many individual organs has been determined.

The purpose of my work is to show the distribution of the water in mammals and to determine whether there are water depots in the body similar to the glycogen and fat depots, and, if such exist, to study a few of the factors influencing the storage or discharge of fluids from them. My purpose is also to determine the relative importance of the different tissues of the body as water depots.

DISTRIBUTION OF WATER IN MAMMALS

All tissues contain water, even the enamel of teeth. Blood, lymph, digestive juices, urine, sweat, feces and expired air are from 80 to 90 per cent water; the organs and softer tissues, about 75 per cent.

Table 1 shows the percentage of water in the body and in the different organs in relation to their weight, for man, rat, dog, cat and rabbit. This table reveals that the water content of the corresponding organs in the different animals is approximately the same. If one should examine the literature cited, one would find that the differences in the water content of the corresponding organs between man and these animals (excepting only a few instances) do not vary more than the figures given by the different authors for the organs of man. Particular attention is called to the small differences between the figures for man, for the cat and for the rabbit. It is also shown that the tissues containing the highest percentage of water are those which are concerned with the transportation of water, and which are under continuous activity, that is, the blood and the heart; also those concerned in the excretory process, namely, the lungs and the kidneys.

The tissues having the lowest percentage composition of water, such as the skeleton and the skin, form the framework and protective covering of the body. The liver also ranks among those being low in water content.

The intestine and skeletal muscle rank intermediate to the other tissues and organs.

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* From the Department of Physiology, University of Minnesota.

TABLE 1.—*Percentage of Water in Organs in Relation to Their Weight*

Organs	Man*	Rat†	Dog‡	Cat§	Rabbit¶
Skin	72.03	77.1	58.9	68.3	71.4
Muscles	75.67	76.2	73.5	74.5	75.08
Skeleton	22.04	47.4	31.66	32.8	20.54
Brain	74.84	77.5	75.03	78	77.93
Spinal cord	.....	69.13	69.53	.....	69.42
Liver	68.25	74	73.7	67.9	75.8
Heart	79.21	77.6	78.39	78.46	78.58
Lungs	78.96	81.6	78.7	76.3	77.65
Kidney	82.68	77.1	76.72	75.3	78.86
Spleen	75.77	77.4	78.6	78.2	80
Blood	83	80.82	82.8	86.7	82.8
Intestine	74.54	76.36	75.4	78.3	78.4
Entire body	63	65.3	65.7	64.2	68.8

* Bischoff, E.: Ztschr. f. rat. Med. **20**: 75, 1863. Engles, W.: Arch. f. exper. Path. u. Pharmakol. **51**: 346, 1904. Herman, L.: Handbuch der Physiologie **6**: 345, 1891.

† Donaldson, H. H.: Memoirs of the Wistor Institute, Philadelphia, 1915.

‡ Engles, W.: Arch. f. exper. Path. u. Pharmakol. **51**: 346, 1904. Stewart, G. N.: Am. J. Physiol. **58**: 45, 1921.

§ Volt, von Carl: Ztschr. f. Biol. **2**: 353, 1866.

¶ Krause, W.: Zwéite Auflage, Leipz., 1884.

TABLE 2.—*Percentage Weight of Organs in Relation to Body Weight**

Organs	Man	Rat	Dog	Cat	Rabbit
Skin	18	18.01	16.11	13.94	13.2
Muscle	41.7	45.4	42.84	45.36	52.2
Skeleton	15.9	10.9	17.39	12.67	12.4
Brain	2.01	0.46	1.37	1.04	0.58
Spinal cord	....	0.16	....	0.07	0.18
Liver	2.26	4.17	3.6	2.96	5.2
Heart	0.47	0.34	0.81	0.37	0.88
Lungs	0.69	0.53	0.88	0.51	1.24
Kidneys	0.37	0.78	0.65	0.81	0.71
Spleen	0.18	0.25	0.22	0.28	0.1
Blood	4.9	5.5	7	4.46	6.75
Intestine	1.81	3.96	6.5	3.8	4
Rest of body	4.63	2.89	2.63	4.1	1.88

* The footnotes for this table are the same as those for table 1, with the exception of those referring to the column on "Man," which are as follows: Bischoff, E.: Ztschr. f. rat. Med. **20**: 75, 1863. Engles, W.: Arch. f. exper. Path. u. Pharmakol. **51**: 346, 1904.

TABLE 3.—*Distribution of Water to Various Organs as Computed from Values Set Forth in Tables 1 and 2 When Body Weights Are: Man, 65 Kg.; Rat, 450 Gm.; Dog, 4 Kg.; Cat, 2 Kg.; Rabbit, 2.5 Kg.*

	Man		Rat		Dog		Cat		Rabbit	
Organs	O. W.	G. W.	O. W.	G. W.	O. W.	G. W.	O. W.	G. W.	O. W.	G. W.
Skin	11,700	8,437.5	81	62.45	644.4	379.5	278.8	190.4	330	235.6
Muscle	27,105	20,510	204	155.6	1,713.6	1,259	907.2	675.8	1,305	979.7
Skeleton	10,335	2,277	49	23.2	695.6	220.2	253.4	83.1	310	63.6
Brain	1,306	977.7	2	1.6	54.8	41.1	20.8	20.5	14.6	11.3
Spinal cord	.....	.....	0.7	0.5	.....	.....	5.4	....	4.7	3.2
Liver	1,469	1,002.5	18.7	13.8	144	106.1	59.2	40.1	130	98.5
Heart	305.5	241.9	1.5	1.1	32.4	25.3	7.4	5.8	22	17.2
Lungs	448.5	354.1	2.3	1.9	35.2	27.7	13.8	10.5	31	24
Kidneys	240.5	198.8	3.5	2.7	26	19.9	7.4	5.5	17.7	13.9
Spleen	117	88.6	1.1	0.9	8.8	6.9	5.6	4.3	2.5	2
Blood	3,195	2,651	24.7	20	280	231.8	89.2	77.3	168.7	139
Intestine	1,176.5	876.9	17.8	13.6	260	196	78	59.5	100	78
Rest of body	.....	6,046	....	6.1	...	114	..	11.4	...	51.3
Total		40,950		293.8		2,628		1,284		1,171

O. W. indicates weight of organ in grams; G. W., grams of water in the organ.

Table 2 is a tabulation of the percentage weights of various organs of man, rat, dog, cat and rabbit in relation to body weight. Attention is again directed to the fact that this weight in the different animals does not vary any more than the figures for any one organ within the same species.

The weights of the different organs and the grams of water each contains in the individual animals of a given weight are tabulated in table 3. This table shows that a man weighing 65 Kg. contains over 40.9 liters of water and that a cat weighing 2 Kg. contains over 1.28 liters, which is distributed to the various organs as shown in table 5.

Examination of tables 3 and 4 shows that nearly half of the water in the body is in the muscle, the most active and most frequently used tissue of the body. The skin contains about one fifth of the water in the body. The great importance of evaporation of water from the skin in maintaining a constant body temperature must be remembered. Although the blood mass represents but 5 per cent of the body weight, it is 82 per cent water and occupies third place among the tissues in water content. It contains, then, in a man weighing 65 Kg., approximately 2.65 liters of water as compared to the 37 to 38 liters of water found in the other tissues.

The water of the blood is not distributed equally between the corpuscles and plasma, which make up from 40 to 60 per cent of whole blood, respectively. I have found plasma to be approximately 92 per cent water and corpuscles 68 per cent.

In table 3 it is seen that a man weighing 65 Kg. has about 3,195 Gm. of blood, of which 2,651 Gm. is water. Of this whole blood, 1,533.6 Gm. are corpuscles, and 1,661.4 Gm. are plasma. From these figures, I find that the corpuscles contain 869.04 Gm., or 33 per cent, of the water in the blood. The plasma contains 1,763.64 Gm. or 67 per cent.

It is this 1.76 liters that gives the blood its fluid consistency and enables it to serve its important function of supplying food, oxygen, and other necessities (internal secretions, hormones and enzymes) to the tissue of the body, and of relieving the other 38 liters of waste material to transport it to the proper localities for its discharge from the body.

It is interesting to note in table 4 that the body water in the different species is distributed similarly to that in man.

PREVIOUS WORK ON WATER DEPOTS OF THE BODY

It is known that after a hemorrhage, water is drawn from the tissues into the blood stream, and that salt solutions injected into the blood stream disappear rapidly.

Although these facts concerning hemorrhage and intravenous injection of salt solution have long been known, no one has made an attempt to determine where the fluid comes from in hemorrhage, and only Engles

has attempted to show to what tissues it goes in the case of injected salt solution. He found that following intravenous injection of physiologic sodium chloride solution, all the tissues except bone became "watery." The muscles, skin and kidneys take up the greater proportion, increasing their percentage amount of water 3.86, 3.23 and 3.83 per cent, respectively. The muscles representing 40 per cent of the body weight take up more than two thirds of any added water, but continue to act normally in spite of the added moisture.

TABLE 4.—*Percentages of Entire Amount of Water in Body Held by Various Tissues as Computed from Values Given in Table 3*

Organs	Man	Rat	Dog	Cat	Rabbit
Skin	20.6	21.2	14.4	14.8	13.7
Muscle	43.4	51.9	47.9	52.6	56.9
Skeleton	5.5	6.9	8.3	6.4	3.7
Brain	2.3	0.56	1.56	1.59	0.63
Spinal cord	...	0.001	....	....	0.18
Liver	2.4	4.72	4.03	3.2	5.72
Heart	0.58	0.4	0.96	0.45	1
Lungs	0.86	0.66	1.05	0.81	1.39
Kidneys	0.48	0.92	0.75	0.43	0.81
Spleen	0.21	0.3	0.26	0.34	0.11
Blood	6.47	6.8	8.82	6.02	8.12
Intestine	2.14	4.63	7.45	5.63	4.55
Rest of body	14.86	1.11	4.44	8.64	3.15

TABLE 5.—*Liters of Water Distributed in the Various Organs in a Man Weighing 65 Kg. and in a Cat Weighing 2 Kg.*

Organs	Man	Cat
Muscle	17.7	0.6
Skin	8.4	0.1
Blood	2.65	0.07
Skeleton	2.27	0.08
Liver	1	0.04
Brain and cord	0.97	0.02
Intestine	0.87	0.05
Lungs	0.35	0.01
Heart	0.24	0.005
Kidneys	0.19	0.005
Spleen	0.08	0.004

In his work, Engles used dogs in series. On one series he made determinations of normal water content; on another, of the water content of the tissues after intravenous injection of physiologic sodium chloride solution. Since muscle constitutes 40 per cent of the body weight and takes up more than two thirds of any added water, he assigns to muscle the greatest importance as water depot.

MY EXPERIMENTS

In my experiments I have determined the water content of a few of the most important tissues before and after altering the water supply on the same animal, in an attempt to show from what tissues most of the water comes after a hemorrhage and to what tissue it goes when injected as salt solutions.

The experiments were carried out on cats in the following manner:

1. Preliminary experimental care of the animal was disregarded except in the first series, in which the alteration of the water balance was effected by hemorrhage. The animals had had (*a*) no previous experimental care, (*b*) no food or fluid for thirty hours previous to the experiment and (*c*) only water for five days.

2. The cats were anesthetized with ether and their weight was recorded.

3. The hair was clipped closely and evenly from the legs, abdomen and neck.

4. A cannula was put in the carotid artery in order to obtain specimens of blood.

5. The abdominal wall was opened, but closed immediately with hemostats in order to prevent the loss of water by evaporation.

6 The gastrocnemius muscle was exposed, and the opening was closed with a hemostat in order to prevent the muscle from losing water by evaporation.

7. The animal was prepared so that all specimens could be taken almost instantaneously. Two samples of each tissue (skin, muscle, spleen, intestine, liver and blood) were placed in weighing bottles that had been accurately weighed. Two samples of each tissue were taken for the purpose of having a check.

8. A sample of blood (2 cc.) for hemoglobin was collected in a small bottle containing a few crystals of potassium oxalate.

9. The kidneys were tied off in order to decrease the loss of water from the body.

10. Alteration of the water balance was made by one of the following procedures:

(*a*) Hemorrhage, 15 cc. per kilogram of body weight.
(*b*) Intravenous injection of distilled water, 25 cc. per kilogram.
(*c*) Intravenous injection of 0.45 per cent sodium chloride, 20 cc. per kilogram.
(*d*) Intravenous injection of 0.9 per cent sodium chloride, 20 cc. per kilogram.
(*e*) Intravenous injection of 1.8 per cent sodium chloride, 20 cc. per kilogram.
(*f*) Intravenous injection of 0.6 per cent calcium chloride, 20 cc. per kilogram.
(*g*) Intravenous injection of 1.2 per cent calcium chloride, 20 cc. per kilogram.
(*h*) Intravenous injection of 2.4 per cent calcium chloride, 20 cc. per kilogram.

11. Thirty minutes were allowed for an effect to take place. All incisions were kept closed with hemostats, and the body was covered with a sheet.

12. Samples of tissue were clipped from as nearly the corresponding place on the other side of the body as possible. Two samples were also taken here for a check.

13. The samples were weighed and put in an electric oven at from 102 to 104 C. until the weight became constant.

14. After cooling the samples in a desiccator over sulphuric acid, the weight was taken, and the percentage of water in each sample was calculated.

The method used in making determinations of hemoglobin consisted in taking the sample of blood collected before the water balance was altered as 100 per cent. The second sample, taken thirty minutes after the water balance was altered, was compared with the normal in a colorimeter. The three-way tap pipet described by Scott[1] was used to insure exactly the same amount of blood in each case.

The technic was so developed by experience that I was able to obtain samples of each type of tissue varying not more than 0.04 Gm. in weight. My next difficulty was to consider the blood content of these tissues.

Naturally, the blood content of these tissues would greatly alter my determinations of water. To overcome this difficulty, I cut the sample in pieces and allowed the blood exuding from each piece to be absorbed by paper toweling. Each sample was treated in exactly the same manner to the minutest detail.

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1. Scott, F. H.: Factors Influencing the Interchange of Fluid Between Blood and Tissue Spaces. I. Blood Pressure, Am. J. Physiol. **44**:298, 1917.

TABLE 6.—*Percentage of Water Contained in the Various Organs Before and After Hemorrhage*

Cat	Weight in Kg.	Skin			Muscle			Spleen			Liver			Intestine			Blood		
		Organ Weight, Gm.	Before	After	Organ Weight, Gm.	Before	After	Organ Weight, Gm.	Before	After	Organ Weight, Gm.	Before	After	Organ Weight, Gm.	Before	After	Organ Weight, Gm.	Before	After
1	1.63	227.2	69.2	68	739.8	77.4	76.8	4.5	78.8	78	48.24	76.7	74.7	61.9	78.9	77.1	72.6	81.1	85.7
2	2	278.8	61.9	60.7	907.2	78.9	78.5	5.6	79.7	79.2	59.20	75.8	74.1	76	78.2	77.1	89.2	85.1	89.5
3	2.5	348.5	67.3	64.1	1,134	75.6	75.2	7	78.6	78.1	74	77	73.9	95	77.3	76.4	111.5	80.2	82.2
4	2.1	292.7	67.6	63.6	952.5	74.9	74.5	5.8	78.7	78.1	62.16	78.2	75	79.8	79	78.4	93.6	81	82
5	1.5	209.1	66.2	63.3	680.4	77.2	76.3	4.2	79.1	78.7	44.4	76.2	73.1	57	78.3	76.6	66.9	81	82.5

	Extent of Hemorrhage in Cc.	Percentage of Hemoglobin After Hemorrhage
1	30	66
2	30	78
3	40	83
4	40	80
5	30	85

TABLE 7.—*Computations Made from Table 6 on the Water Content of Various Organs Before and After Hemorrhage*

Cat*	Skin			Muscle			Spleen			Liver			Intestine			Blood		
	B.	A.	G.D.	B.	A.	G.D.	B.	A.	G.D.	B.	A.	G.D.	B.	A.	G.D.	G.W.	L.T.	%
1	157.2	154.4	— 2.8	572.6	568.1	—4.5	3.59	3.55	—0.04	37	36	—1	48.87	47.72	—1.15	30.1	9.49	31.5
2	172.5	169.2	— 3.3	607.6	604	—3.6	4.46	4.43	—0.03	44.8	43.8	—1	59.40	58.4	—1	25.9	6.63	25
3	234.5	223.3	—11.2	857.3	852.7	—4.6	5.50	5.40	—0.10	56.9	54.5	—2.4	73.40	72.5	—0.90	28.8	19.20	66
4	197.8	186.1	—11.7	713.4	709.6	—3.8	4.60	4.50	—0.1	48.6	46.6	—2	63	62.5	—0.50	23.8	18.10	76
5	138.4	134.4	— 4	525.2	519.1	—6.1	3.32	3.31	—0.01	33.8	32.4	—1.4	44.60	43.6	—1	13.5	12.5	92

B indicates grams of water in the organ before hemorrhage; A, grams of water in the organ after hemorrhage; G. D., grams of water taken up or lost by organ; +, taken up by tissue; —, lost to tissue; G. W., grams of water gained by blood; L. T., total grams of water given up by tissues; %, percentage of fluid entering the blood stream found to be coming from the tissues examined.

* Cats 1 and 2 were normal from the stockroom (typical of ten such experiments); cats 3 and 4 had no food or fluid for thirty-two and seventy-two hours, respectively (typical of six such experiments); cat 5 had only water for five days (typical of four such experiments).

The next difficulty to overcome was that of obtaining dry tissue. Because it is so highly hydroscopic, Benedict expressed his doubt, in a personal conversation, as to whether any one had ever obtained dry tissue. I dried a few samples of tissue in an electric oven from 102 to 104 C. over a period of from twenty-four to thirty-six hours.

Samples of muscle weighing 0.183 Gm. immediately after removing from the oven increased in weight by 0.024 Gm. or 24 mg. when exposed to the air for five minutes. When exposed in a desiccator for the same length of time, a sample of muscle weighing 0.2 Gm. increased its weight by 0.016 Gm. or 16 mg. Other samples dried in the oven at the same temperature and over the same time did not gain in weight if the stopper was placed on the weighing bottle before removing it from the oven. Some of these samples were exposed in the air and others were left in the desiccator for a week without undergoing any noticeable change.

Believing these difficulties to be overcome, I proceeded with the control experiments, which consisted of carrying out the experimental procedure as just outlined, eliminating only the factor altering the water balance. Results from a series of five control experiments show that the percentage of water in the tissues examined did not vary more than 0.2 of 1 per cent at the most in any case. I considered 0.3 per cent as the limit of error.

After establishing the reliability of my procedure and methods, I proceeded with the experimental work, the results of which are shown in the accompanying tables; those for hemorrhage are given in detail to show the type of data used; those for the injections of watery solutions are summarized in table 10.

COMMENT

Table 7 shows that severe hemorrhage resulted in the blood becoming more "watery," while the other tissues of the body lost water. This result was true whether the animal was starved, given only water or taken direct from stock before the experiment.

Tables 8 and 9 show the average results. Although the greatest amount of water entering the blood stream after hemorrhage in normal cats from stock (cats 1 and 2) comes from the muscles, they lost the least amount (0.5 Gm.) of water for each hundred grams of tissue. This is also true of the results obtained with cat 5 (water only for five days). In this case 45.2 per cent of the water entering the blood stream came from the muscles at the rate of 0.9 Gm. for each hundred grams of tissue. In these same experiments, the next greatest amount (11 per cent) of water came from the skin. The liver contributed approximately the same percentage (3.5 per cent) as the intestine (3.6 per cent) in cats 1 and 2, but gave a much larger quantity in the experiment with cat 5.

Further study of these experiments shows that for each hundred grams of tissue the liver lost the greatest amount of water (2.1-3.2 Gm.), while the intestine and the skin lost approximately the same amount (1.6 and 1.2 Gm.).

These results bear an analogy with Starling's [2] interpretation of the relative permeability of capillaries in different parts of the body. He found that lymph from muscles of the limbs was from 2 to 4 per cent protein; that from the intestine, from 4 to 6 per cent; that from the liver, from 6 to 8 per cent. From this he deducted that the capillaries of the liver were the most permeable, those of the intestine next and those of the muscle the least. This parallels the results already mentioned;

TABLE 8.—*Source of Fluid Entering Blood Stream Expressed in Percentage of Amount Coming from Various Tissues*

Cat	Skin	Muscle	Liver	Intestine	Spleen
1	9.3	14.9	3.3	3.4	0.13
2	12.7	14	3.8	3.8	0.03
Average	11	14.45	3.5	3.6	0.08
3	38	16	8.3	3.1	0.34
4	49	16	8.4	2.1	0.42
Average	43.5	16	8.35	2.6	0.07
5	30	45.2	10.4	0.74	0.72

TABLE 9.—*Amount of Fluid Given Up by 100 Gm. of Tissue Following Hemorrhage Computed from Table 6*

Cat	Skin	Muscle	Liver	Intestine	Spleen
1	1.2	0.6	2.2	1.9	1
2	1.2	0.4	2	1.3	0.6
Average	1.2	0.5	2.1	1.6	0.8
3	3.2	0.4	3.2	0.9	3.3
4	4	0.39	3.2	0.7	2.17
Average	3.6	0.4	3.2	0.8	2.73
5	2.1	0.9	3.2	1.75	0.25

that is, that muscle, with the least permeable capillaries, lost the least water per unit mass of tissue, and that the liver, with the most permeable, lost the most per unit mass.

An average of the results from cats 3 and 4 (no food or fluid for thirty-two and seventy-two hours, respectively) shows that the skin contributed not only the greatest amount (3.6 Gm.) for each hundred grams of tissue, but also a greater percentage (43.4 per cent) of the fluid entering the blood stream than any of the other tissues. This was not only an increase over the amount lost for each unit mass of tissue in cats 1 and 2, but also an increase in the percentage of water it contributed toward that entering the blood stream.

The next greatest percentage (16 per cent) came from the muscles, but at the rate of only 0.4 Gm. per hundred grams of tissue. This was

2. Starling, E. H. The Influence of Mechanical Factors on Lymph Production, J. Physiol. **16**:224, 1894.

TABLE 10.—*Changes in the Water Content in Various Organs Following Intravenous Injection of Watery Solutions*

	Distilled Water		0.45% Sodium Chloride		0.9% Sodium Chloride		1.8% Sodium Chloride		0.6% Calcium Chloride		1.2% Calcium Chloride		2.4% Calcium Chloride	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Blood	23.7	17	37.26	20	49.45	27.7	59.3	33.7	64.4	36.1	74.5	41.8	100+	
Muscle	13.1	0.75	21.70	1.3	0	0	Loss	—1.2	16.4	1.1	Loss	—4.3	Loss	—0.5
Liver	9.5	7.6	5.57	4.6	3.91	4.3	3.45	4.1	5	3.0	3.2	2.4	Loss	—2
Skin	5.8	1.1	11.1	1.9	13.57	4.5	17.5	4.8	8.1	1.14	Loss	—3.55	Loss	—4.2
Intestine	2.7	2.1	4.5	2.8	2.21	1.9	2.81	1.9	2.6	1.33	3.9	2.6	Loss	—0.6
Spleen	0.16	1.6	0.09	0.9	0.07	0.9	0.09	0.9	0.18	0.87	0.26	2.4	0	0

Under A are listed the percentage of the injected fluid remaining in the blood stream or absorbed by the various tissues at the end of thirty minutes; under B, the grams of fluid taken up or lost (—) by 100 Gm. of tissue.

The results with distilled water are an average of six experiments: with 0.45% sodium chloride, 8; 0.9% sodium chloride, 8; 1.8% sodium chloride, 6; 0.6% calcium chloride, 6; 1.2% calcium chloride, 6; 2.4% calcium chloride, 4.

by far the least amount of water lost per unit mass by any tissue in these experiments.

Of the total amount of fluid entering the blood stream, 8.35 per cent came from the liver at the rate of 3.2 Gm. per hundred grams of tissue. This was an increase of 1 Gm. of water for each hundred grams of tissue over the results with cats 1 and 2. It is nearly as much as for each unit mass of skin, and half as much as for each unit mass of muscle. Here one must recall that there is approximately fifteen times as much muscle as liver in the cat, and yet muscle lost only twice as much water as the liver.

The intestine ranks fourth in regard to the amount (2.6 per cent) of water that came from it after the hemorrhage. For each hundred grams of tissue it lost 0.8 Gm. of water, being 0.3 of a Gm. more than that lost by muscle.

For each hundred grams of its tissue the spleen lost 2.73 Gm. of water, but because of its size, only 0.07 per cent of the fluid that entered the blood came from it.

Examination of table 10 shows that distilled water leaves the blood stream more rapidly than any of the other solutions. Solutions having a higher salt content left the blood more slowly, and the increase in water content shown by the tissues decreased. With isotonic sodium chloride there was no change in the water content of muscle, but there was a decrease in its water content thirty minutes after the injection of hypertonic sodium chloride. Following the injections of hypotonic calcium chloride, the tissues did not show as big an increase in their water content as they did when hypotonic sodium chloride was injected. With the exception of muscle, the tissues did not become as "watery" as they did after the injection of isotonic or even of hypertonic sodium chloride. There was also a greater percentage of the injected solution remaining in the blood stream at the end of thirty minutes than there was when hypertonic sodium chloride was injected.

Only 26 per cent of the isotonic calcium chloride solution injected had left the blood stream at the end of thirty minutes. Muscle and skin both showed a decrease in their water content after its injection.

Following the injection of a 2.4 per cent solution of calcium chloride, there was more water (above normal amount) in the blood stream at the end of thirty minutes than had been injected. The principal source for this water was the skin, and the next most important source was the muscle; some came from the intestine, but little came from the spleen.

Following the injection of distilled water or the hypotonic salt solutions, the increase in the water content of the tissues as expressed in grams of water taken up by 100 Gm. of tissue is parallel to Starling's interpretation of the permeability of the capillaries in those regions. The liver showed the greatest increase for each hundred grams of its

tissue, while the muscle showed the least. It is worthy of note, however, that muscle took up a larger percentage of the water added per se than any other tissue. Liver took up the next greatest amount; skin, the third greatest amount; the intestine, the fourth largest amount and spleen the least.

This was relatively true when hypotonic salt solutions were injected, except that skin not only showed an increase of more per unit mass of tissue than it did with distilled water, but it also absorbed a greater percentage of the injected fluid than did the liver. When isotonic sodium chloride was injected, the skin not only showed an increase of more water per unit mass of tissue than it did following the injection of the hypotonic solution, but also contained a greater percentage of the fluid injected than any other tissue. Likewise, following the injection of a hypertonic sodium chloride solution, the skin showed a still greater increase in water content per unit mass of tissue than it did with isotonic sodium chloride; there was also an increase in the percentage amount of the injected fluid that it had taken up.

The injection of isotonic sodium chloride solution did not result in an alteration of the water content of muscle, but after the injection of a hypertonic solution there was a diminution in its water content.

This result differs from that of Engles,[3] who found that muscle increased its water content by 3.86 per cent following the injection of physiologic sodium chloride. His work was carried out on dogs in series and not as a complete experiment on one animal.

In his work on the significance of the tissues as a depository for chlorine, Wahlgren[4] found that the muscles not only had the lowest percentage of chlorine content, but also increased their percentage amount the least when chlorine was given intravenously in the form of sodium chloride. He also found that water passed from the muscles into the blood after intravenous infusion of hypertonic sodium chloride solutions.

The results of Tashiro,[5] who observed the effect of injecting hypotonic and hypertonic saline on the water content of striated muscle of rabbits obtained the same results as I did on that tissue.

Injection of 1.2 per cent calcium chloride resulted in a marked decrease in the water content of the skin and muscle, but in an increase in the water content of the liver, intestine and spleen. The muscle lost more (4.3 Gm.) for each hundred grams of its tissue than the skin. The

3. Engles, W.: Die Bedeutung der Gewebe als Wasserdepots, Arch. f. exper. Path. u. Pharmakol. **51**:346, 1904.

4. Wahlgren, V.: Ueber die Bedeutung der Gewebe als Chlordepots, Arch. exper. Path. u. Pharmakol. **61**:97 (Sept. 30) 1909.

5. Tashiro, N.: Changes in Water Content of Striated Muscles Caused by Disturbances of Water Metabolism of Rabbits, Arch. f. exper. Path. u. Pharmakol. **3**:218, 1926.

intestine showed an increase of 2.6 Gm. of water for each hundred grams of tissue as compared to 2.4 Gm. for each hundred grams of tissue in the liver and in the spleen.

There was a marked depletion in the water content of all the tissues following the injection of 2.4 per cent calcium chloride solution. The effects of this injection parallel those observed after severe hemorrhage. The blood became more "watery" than could be accounted for by the fluid injected, and all tissues lost water. The muscle lost the least per unit mass of tissue and, with the exception of skin, the liver lost the most.

SUMMARY AND CONCLUSIONS

1. The distribution of water in man and in the rat, dog, cat and rabbit given in table 4 shows that approximately half of the body water is in the muscles. The skin contains about one fifth of the water in the body, and the blood about 7 per cent. Of the water in the blood, 67 per cent is in the plasma and 33 per cent is in the corpuscles.

2. All the tissues lost water after hemorrhage. The amount of water entering the blood came from the organs in the following order (tables 8 and 9):

A. In cats having a normal supply of water in their tissues: (1) muscle, 14.45 per cent; skin, 11 per cent; intestine, 3.6 per cent; liver, 3.5 per cent; spleen, 0.08 per cent. (2) Per hundred grams of tissue, the order was as follows: liver, 2.1 Gm.; intestine, 1.6 Gm.; skin, 1.2 Gm.; spleen, 0.8 Gm.; muscle, 0.5 Gm.

B. In cats that had been deprived of water and food for five days: (1) skin, 43.5 per cent; muscle, 16 per cent; liver, 8.35 per cent; intestine, 2.6 per cent; spleen, 0.07 per cent. (2) Per hundred grams of tissue: skin, 3.6 Gm.; liver, 3.2 Gm.; spleen, 2.73 Gm.; intestine, 0.8 Gm.; muscle, 0.4 Gm.

3. Following the injection of distilled water or hypotonic salt solutions, the fluid leaving the blood stream at the end of thirty minutes was distributed to the tissues in the following order (table 10): (1) muscle, 13-21 per cent; liver, 9.5-5.5 per cent; skin, 5.8-11 per cent; intestine, 2.7-4.5 per cent; spleen, 0.1-0.8 per cent. (2) Per hundred grams of tissue, the amount added was as follows: liver, 7.6-3 Gm.; intestine, 2.1-1.3 Gm.; spleen, 1.6-0.8 Gm.; skin, 1.1-1.9 Gm.; muscle, 0.7-1.1 Gm.

4. Following the injection of isotonic and hypertonic sodium chloride solutions all tissues, except muscles, showed an increase in their water content.

A. After isotonic sodium chloride, there was no change in the water content of muscle, while there was a decrease in its content after hypertonic sodium chloride was injected.

B. The skin showed a greater increase in its water content the greater the sodium chloride content of the injected solution. Its increase per unit mass of tissue was least after distilled water and greatest after hypertonic sodium chloride was injected.

5. Following the injection of 1.2 per cent calcium chloride solutions, the muscle and skin showed a decrease in their water content. Muscle showed the greatest difference per unit mass of tissue. The intestine showed an increase of 2.6 Gm. of tissue as compared to 2.4 Gm. for each hundred grams of tissue in the liver and of the spleen.

6. The results after injecting 2.4 per cent calcium chloride were parallel to those after hemorrhage.

7. Although the muscle contains nearly one half of the body water, and loses the least per unit mass of tissue, it gives up more fluid than any other tissue when the animal is deprived of fluid, and takes up by far the greatest portion of any water added as water per se or as a hypotonic salt solution. It would seem justifiable, therefore, to indicate muscle as the most important water reserve of the body; it stores the greatest quantity of any excess, and acts as a safeguard against the loss of too much water from the body.

8. The importance of skin as a reserve to take care of any excess fluid, especially when added as a sodium chloride solution, must not be overlooked. The experience with partially dehydrated animals indicates that skin will furnish the greatest quantity of water to the body if a further dehydration should occur.

9. The liver and the intestine appear to respond more quickly than any other tissues when there is an alteration in the water content of the body.

10. The promptness and the degree of reaction of the various tissues examined to any alteration in the fluid content of the body bear a relation to Starling's interpretation of capillary permeability. This is particularly true in the case of hemorrhage, or when distilled water or hypotonic salt solutions are injected.

RELATIVE HYPOTENSION OF FOREIGNERS IN CHINA*

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It has been the common experience in studying groups of healthy Chinese to find that the blood pressure, both systolic and diastolic, is lower than that in occidental races. Table 1 summarizes the results of several investigations.

In the West, with special reference to the United States, Alvarez [1] found that the average systolic blood pressure of 6,000 healthy men, ranging in age from 16 to 40 years, was 129 millimeters of mercury and in women 117. From a group of 141,840 persons of both sexes, ranging in age from 20 to 44 years, Symonds [2] obtained an average systolic blood pressure of 125 and a diastolic pressure of from 80 to 84. According to

TABLE 1.—*Records of Blood Pressure of Chinese Men*

Author	Locality	Number of Subjects	Age	Average Systolic Pressure, Mm. Hg	Average Diastolic Pressure, Mm. Hg
Kao (Nat. M. J. China **8**: 101, 1922).....	Hunan..... (Central China)	63	21 to 25	116	71
Kao (Nat. M. J. China **8**: 101, 1922).....	Hunan..... (Central China)	10	26 to 42	113	71
Cadbury (China M. J. **37**: 823, 1923).....	Canton..... (South China)	700	15 to 30	101	65
Kilhorn (China M. J. **40**: 1, 1926).....	Szechuan..... (West China)	741	14 to 31	111	70
Ying (China M. J. **40**: 641, 1926).....	Shaoshing..... (East China)	182	21 to 50	113	72

the available evidence, therefore, the average systolic pressure of healthy adults in the United States is approximately 125 and the diastolic 80, with slightly lower figures (from 5 to 10 lower) for women, while the average systolic pressure of Chinese is about from 110 to 115 and the diastolic about 70.

Although both the systolic and the diastolic blood pressure of the Chinese is known to be about 10 lower than that of westerners, little study has been made of the effect of residence in the Orient on the blood pressure of westerners. Musgrave and Sison [3] were the first to point out an apparent lowering of blood pressure following residence in the Philippines for from one to ten years or more. Their figures, based on the blood pressure of ninety-seven foreigners (varying in age from

* From the Department of Medicine, Peking Union Medical College.

1. Alvarez, W. C.; Wulzen, R., and Mahoney, L. J.: Blood Pressure in 15,000 University Freshmen, Arch. Int. Med. **32**:17 (July) 1923.

2. Symonds, B.: The Blood Pressure of Healthy Men and Women, J. A. M. A. **80**:232 (Jan. 27) 1923.

3. Musgrave and Sison: Philippine J. Sc., Sec. B. **5**:325, 1910.

25 to 40), most of whom were Americans in the Philippine Civil Service and the United States Army, were as given in table 2 (Erlanger instrument used).

The figures for ten French Sisters of Charity (from 24 to 38 years), were as given in table 3.

These figures show that apparently there is a progressive decrease in blood pressure accompanying prolonged sojourn in the tropics.

In 1911, Chamberlain [4] found that the average systolic pressure of 992 American soldiers serving in the Philippines (average age, 26) was 115. The average length of their term in this service was about eleven months. This figure coincided with the average systolic pressure of 115 in 386 Filipinos whose average age was 25 years. Chamberlain concluded that for neither race is the blood pressure materially below the

TABLE 2.—*Average Blood Pressure of Foreigners in the Philippine Islands*

Length of Residence	Average Systolic Pressure
1 month to 1 year (65 cases)	124
1 year to 5 years (15 cases)	115
5 years to 10 years (8 cases)	116
Over 10 years (9 cases)	113

TABLE 3.—*Average Blood Pressure of Ten French Sisters of Charity*

Length of Residence	Average Systolic Pressure
15 days to 1 year	127
1 year to 5 years	112
Over 5 years	110

figures to be expected for white men residing in the temperate climates. This conclusion may be questioned for two reasons: First, the average systolic pressure of healthy men, aged 26, in the West is about 125, as has just been shown; second, the period of residence in the tropics in this author's cases was probably too short to have any marked effect on the blood pressure. Foster [5] reported that the blood pressure of foreigners in Changsha was about the same as that of the Chinese, and that the blood pressure of many persons was lower in China than it was in America. He based the latter statement on about thirty records taken both in America and in Changsha.

In table 4 I have compiled the blood pressure readings, systolic and diastolic, of fifty-eight westerners (mostly Americans), members of the staff of the Peking Union Medical College, showing determinations

4. Chamberlain: Philippine J. Sc., Sec. B. **5**:467, 1911.

5. Foster: Abstracts of Papers for China Medical Association Conference, Peking, 1926, p. 10.

made at home and in Peking. The age column gives the age of each person at which his blood pressure was first taken. The readings in America or, rarely, in Europe are taken from records of physical examinations made abroad, as indicated, and kept on file here. In about two thirds of the cases, the records in Peking represent those obtained during routine physical examinations performed every two years after the person's arrival in China, or by special appointment for the determination of blood pressure. For the sake of clearness, the results of only the most recent of these examinations are presented in this article. In the other

TABLE 4.—*Parallel Records of Blood Pressure of Fifty-Eight Americans in the United States and in China*

			Blood Pressure in the United States		Blood Pressure in China		Change in Blood Pressure	
Number	Sex	Age	Systolic	Diastolic	Systolic	Diastolic	Systolic	Diastolic
1	M	30	135	75	122*	70*	−13	− 5
2	M	33	120	80	118	65	0	−15
3	M	32	112	76	110	64	0	−12
4	M	39	120	70	110	60	−10	−10
5	M	35	128	70	110	70	−18	0
6	M	30	118	80	125	75	+ 7	− 5
7	M	44	122	80	110	70	−12	−10
8	M	34	120	76	114	75	− 6	0
9	M	29	120	90	110	66	−10	−24
10	M	24	100	70	106	64	+ 6	− 6
11	M	36	115	68	120	60	+ 5	− 8
12	M	32	112	78	100	58	−12	−20
13	M	35	130	80	115	75	−15	− 5
14	M	56	110	74	92	58	−18	−16
15	M	44	106	70	112	70	+ 6	0
16	M	39	135	90	108	66	−27	−24
17	M	25	122	80	108	62	−14	−18
18	M	42	125	80	108	66	−17	−14
19	M	37	145	80	84	60	−61	−20
20	M	52	140	105	108	70	−32	−30
21	M	42	120	84	98	56	−22	−28
22	M	45	125	80	110	70	−15	−10
23	M	21	100	60	90	60	−10	0
24	M	22	110	70	138	80	+20	+10
25	M	28	124	82	138	90	+14	+ 8
26	M	23	110	65	108	64	0	0
27	M	38	100	60	112	72	+12	+12
28	M	28	110	75	102	58	− 8	−17
29	F	31	110	80	108	60	0	−20
30	F	40	132	80	110	68	−22	−12
31	F	36	120	80	100	60	−20	−20
32	F	36	104	64	104†	60†	0	0
33	F	33	120	65	110	90	−10	+25
34	F	28	120	80	110	65	−10	−15
35	F	39	118	70	120	70	0	0
36	F	33	115	75	110	58	− 5	−17
37	F	30	105	75	100	50	− 5	−25
38	F	33	115	60	112	70	0	+10
39	F	28	118	84	109	62	− 9	−22
40	F	30	130	80	110	68	−20	−12
41	F	46	108	60	105	65	0	+ 5
42	F	28	100	55	110	55	+10	0
43	F	26	118	82	90	60	−28	−22
44	F	32	115	70	112	65	0	− 5
45	F	34	112	80	106†	66†	− 6	−14
46	F	32	124	80	102	72	−22	− 8
47	F	35	120	76	100	65	−20	−11
48	F	42	121	70	114	78	− 7	+ 8
49	F	28	130	90	110	60	−20	−30
50	F	35?	118	85	118	68	0	−17
51	F	36	106	68	120	60	0	− 8
52	F	45	120	75	98	66	−22	− 9
53	F	29	125	95	114	60	−11	−35
54	F	33	110	60	110†	40†	0	−20
55	F	29	115	85	95	54	−20	−31
56	F	28	115	80	95	60	−20	−20
57	F	51	120	80	130	78	+10	0
58	F	36	130	90	110	70	−20	−20

* Blood pressure in China during diarrhea.
† Blood pressure in China during pregnancy.

third of the cases the readings are taken from histories of patients who have called at the office on account of minor ailments, or from hospital histories. Care has been exercised to exclude readings taken while the patients had diarrhea, dysentery, acute respiratory infections, prolonged fever or illness of any kind, or when patients were pregnant, except in a few cases designated in the column under "Notes."

The average age in the foreign group reported was 34. The time at which the Peking reading was taken is not indicated, because in many cases an average of the readings of different years is presented (in hospital cases), but the average time was from two and one-half to three years after the reading taken abroad. No Peking figure was taken within one year after the person's arrival in China, except in case 8 of the foreign group and in case 3 of the Chinese group, both being obtained within two months after the subject's arrival in China.

In regard to the technic of blood pressure reading, I do not know how the blood pressure in America was taken; in China, it was taken by several different physicians. The apparatus used was a mercury manometer. About half of the blood pressures were taken when the

TABLE 5.—*Grouping of Blood Pressure of Fifty-Eight Americans in the United States and in China*

Systolic pressure.....	80-89	90-99	100-109	110-119	120-129	130-139	140-149
United States.....	0	0	9	20	20	7	2
China.....	1	7	16	25	4	3	0
Diastolic pressure.....	**40-49**	**50-59**	**60-69**	**70-79**	**80-89**	**90-99**	**100-109**
United States.....	0	1	10	18	23	5	1
China.....	1	8	30	16	1	2	0

subjects were in a supine position and half when they were seated. The auscultatory method was used, and the first appearance of a sharp thumping sound was recorded as the systolic pressure; the sudden change from a sharp tap to a dull, feeble thud was regarded as the diastolic pressure.

In the column under "Changes in Blood Pressure," an increase of 5 or more is indicated by a plus sign and a decrease of 5 or more by a minus sign. No change (a change of 4 or less is considered as no change) is signified by "0."

Table 5 shows the grouping of the records of the blood pressure of this group in the United States and in Peking, and no further comment is necessary.

As will be seen from table 6, the average systolic pressure of fifty-eight foreigners in America is 118 and the average diastolic, 76; while the average systolic pressure in Peking is 109 and the average diastolic, 65, although the subjects have increased in average age from about 34 to 37. There is an average lowering of 9 in the systolic, and of 11 in

the diastolic, pressure. Thus it appears that there is a distinct tendency for the blood pressure of foreigners resident in China to approach that of the Chinese.

Analyzed from the point of view of percentage of increase or decrease in blood pressure, it is seen from the results in table 6 that the systolic pressure shows a decrease in 64 per cent, and an increase in only 15 per cent in China. The diastolic pressure is even more striking, with 72 per cent showing a fall in pressure and only 12 per cent a rise.

COMMENT

Although this series is too small to afford a basis for any far-reaching conclusions, it indicates that there is a fall in blood pressure, both systolic and diastolic, in the majority of foreigners who take up their residence in China for any length of time. Musgrave and Sison [3] thought that the decrease in blood pressure was due to a lower peripheral resistance, associated with increased secretory activity of the sweat glands. Gastrointestinal disturbances were also suggested by these authors as capable

TABLE 6.—*Average Blood Pressure in the United States and in China, and Percentage of Observations Showing Changes*

	Fifty-Eight Americans	
	Systolic	Diastolic
United States	118	76
China	109	65
No change	21 per cent	16 per cent
Increase	15 per cent	12 per cent
Decrease	64 per cent	72 per cent

of producing motor disturbances, principally through the portal circulation and the splanchnic tone. It is claimed by Huggard [6] that the low blood pressure in persons who reside in the tropics is associated with dilatation of the peripheral blood vessels, as well as with increased elasticity of the vessel wall.

While the low blood pressure of the Chinese may be due in small part to a diet less rich in meat, the diet of foreigners is not essentially different from that in America. The perpetual rush, tension and excitement that characterize American life and the comparatively slower and calmer life in Peking, although the work here may be no less strenuous, may be important factors in the causation of the relative hypotension. In a recent physiologic review,[7] MacWilliam says: "The pronounced effects of mental stress, excitement and worry in producing and maintaining high blood pressure emphasize the significance of the nervous system

6. Huggard, quoted in Castellani and Chalmers: Manual of Tropical Medisine, ed. 2, 1913, p. 89.

7. MacWilliam: Physiol. Rev. **5**:303, 1925.

whether exercised indirectly through the cardiovascular innervation or more directly through endocrine or metabolic alterations." Lombard [8] and Krogh [9] emphasize the independent contractility of the capillaries and arterioles. Studies on the vasomotor condition in the tropics and in the Orient are lacking at present.

It is true that there is increased sweating in the tropics, but there is no definite knowledge of the relation of sweating to vasomotor states. Evidently there may be a cutaneous vasodilatation associated with sweating, as may be seen during flushing caused by excitement or embarrassment; but there may be also a state of pallor with sweating, as that occurring during fright. In any case this mechanism does not explain the hypotension reported here, since Peking is neither tropical nor subtropical. It is a point of interest that hypertension, which has been considered as bearing a special relation to tropical residence, is also present in the temperate zone.

MacWilliam [7] has epitomized the present knowledge of hypotension:

> But in persistent low pressure attendant on exhausting diseases or occurring without obvious cause (essential hypotension) the available data are, as in the case of persistent high pressure, inadequate for satisfactory explanation of the mechanism involved—whether a defective peripheral resistance or defective cardiac output depending, apart from cardiac enfeeblement, on lessened return of blood to the heart as a result of undue expansion of the capacity of the vascular system from capillary or venous relaxation, contraction of venules, diminished volume of blood in circulation, etc. It is also unknown how far such conditions are mediated through the nervous system and how far due to the direct influence of chemical agents—depressor bodies, lack of pressor substances, etc.

What true significance the relative hypotension of the westerners in China and in the tropics has remains to be seen.

SUMMARY

1. An analysis of parallel records of the systolic and the diastolic blood pressure of fifty-eight Americans at home and in Peking about three years later shows an increase of systolic pressure in 15 per cent of the subjects, no change in 21 per cent and a decrease in 64 per cent. The diastolic pressure shows an increase in 12 per cent, no change in 16 per cent and a decrease in 72 per cent.

2. The average systolic pressure of fifty-eight Americans in America is 118 millimeters of mercury and in China 109. The average diastolic pressure in America is 76, while it is only 65 in Peking. Thus, there is an average decrease of 9 in systolic pressure in Peking and an average decrease of 11 in diastolic pressure.

8. Lombard: Am. J. Physiol. **24**:335, 1912.

9. Krogh: Anatomy and Physiology of the Capillaries, 1922, p. 23.

DISEASES OF THE LIVER

VI. A COMPARATIVE STUDY OF CERTAIN TESTS FOR HEPATIC FUNCTION IN CASES OF CIRRHOSIS OF THE LIVER *

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During the last three years we have been engaged in an attempt to evaluate the clinical usefulness of certain tests for hepatic function. In previous communications we have reported the results of various aspects of this study.[1] At present we wish to report on the functional changes observed in patients with cirrhosis of the liver.

Of a group of more than 100 cases originally diagnosed cirrhosis of the liver, the history and physical observations were characteristic and the diagnosis apparently unmistakable in the fifty cases selected for this report. In the majority the diagnosis was confirmed by examination of the liver at operation or necropsy. Syphilis was excluded in these cases by the history and physical examination and by serologic tests. Observations made in the cases of cirrhosis associated with splenic anemia (so-called Banti's disease) and with hemolytic jaundice have previously been reported.

The presentation of this material has proved singularly difficult because of the present confusion with regard to the classification of cirrhosis. While textbooks of medicine recognize certain types of cirrhosis and make clear-cut distinctions along clinical, etiologic or pathologic lines, any one observing and studying such cases in any number quickly recognizes the great variation in clinical types, and appreciates how incomplete and unsatisfactory is the present knowledge of hepatic disease. Epplen [2] has recently pointed out the confused and unsatisfactory

* From the Division of Medicine, Mayo Clinic and the Mayo Foundation.

1. Greene, C. H.; Snell, A. M., and Walters, W.: Diseases of the Liver: I. A Survey of Tests of Hepatic Function, Arch. Int. Med. **36**:248 (Aug.) 1925. Greene, C. H.; McVicar, C. S.; Rowntree, L. G., and Walters, W.: Diseases of the Liver: III. A Comparative Study of Certain Tests for Hepatic Function in Patients with Obstructive Jaundice, Arch. Int. Med. **36**:418 (Sept.) 1925; Diseases of the Liver: IV. Functional Tests in Cases of Carcinoma of the Liver and Biliary Tract, Arch. Int. Med. **36**:542 (Oct.) 1925. Greene, C. H., and Conner, H. M.: Diseases of the Liver: V. A Comparative Study of Tests of Hepatic Function in Certain Diseases of the Hematopoietic System, Arch. Int. Med. **38**:167 (Aug.) 1926.

2. Epplen, F.: The Pathology of Cirrhosis of the Liver; An Historical-Pathologic Study, Arch. Int. Med. **29**:482 (April) 1922.

nature of the various attempts at classification on an etiologic or pathologic basis. We have felt that an attempt at the reclassification of the cirrhoses at the present time would be premature, of questionable value, and would involve considerable presumption on our part. Nevertheless, presentation of the functional studies demands some recognition of the various types of cases. Therefore we have adopted the following grouping for this purpose:

Portal cirrhosis or ascitic cirrhosis (cirrhosis with ascites, chronic hepatitis with ascites): (1) portal cirrhosis with ascites and small liver, (2) portal cirrhosis with ascites and large liver, (3) portal cirrhosis with ascites and jaundice and (4) large liver without ascites, presumably portal cirrhosis in the preascitic stage.

Biliary cirrhosis or icteric cirrhosis (cirrhosis with chronic jaundice, chronic hepatitis with jaundice): (1) biliary cirrhosis with extrahepatic obstruction, (2) biliary cirrhosis without extrahepatic obstruction and (3) biliary cirrhosis with ascites.

While we may speak of these groups as separate entities, it must be understood that these divisions are made for convenience in presentation and so are necessarily somewhat artificial. Detailed discussion, particularly of the etiology and pathology of the condition, will be reserved for later communications. The complete laboratory data of the clinical investigations are given in the accompanying tables. Abstracts of the histories of illustrative cases are also included. In these only significant laboratory data are given. The phenoltetrachlorphthalein test is reported in terms of one hour reading.

PORTAL CIRRHOSIS WITH ASCITES AND SMALL LIVER ("LAENNEC'S CIRRHOSIS")

In his "Traite de l'auscultation mediate," Laennec [3] gave an anatomic description of cirrhosis which is classic:

> The liver, reduced to a third of its ordinary size, was buried, if I may use the term, in the region which it occupies, and its external surface, slightly pursed and wrinkled, was of a yellowish-grey tint. On incision, it appeared entirely composed of a multitude of small round or ovoid grains, varying in size from that of a millet to that of a hemp-seed. There was scarcely any interval between these grains, in which they could still be distinguished from remains of the proper tissue of the liver; they were easily separable from each other, of a fawn or red-yellow color, inclining in some places to green, and of a humid, opaque tissue, flaccid rather than soft to the touch, so that on pressing the grains between the fingers a small part only crumbled away, the rest feeling in the hand like a piece of soft leather.

Rigid definition would require that the use of the term Laennec's cirrhosis be restricted to those cases of portal cirrhosis in which the liver

3. Laennec, R. T. H.: Traité de l'auscultation médiate et des maladies des poumons et du coeur, ed. 2, Paris, J. S. Claudé, 1826, vol. 2, p. 196; A Treatise on Mediate Auscultation and on Diseases of the Lungs and Heart, London, H. Baillere, 1846, p. 436 (edited by T. Herbert).

was reduced in volume and would exclude those cases in which that organ was increased in size. Such a distinction is of questionable clinical or pathologic value. We mention it here because of its historical association.

The disease usually develops slowly and insidiously. There may be moderate initial nausea or bloating and dull pain or discomfort in the right hypochondrium. Occasionally slight jaundice, the "signal jaundice" of Rolleston,[4] may be present at the time of onset. Usually these symptoms are so trifling that they are passed over unnoticed, and attention is not called to the condition until signs of altered circulation appear. The abdomen then becomes distended and fluctuant. The quantity of fluid may be so great that satisfactory palpation of the abdomen is precluded. When the fluid is removed, the liver may be found to be reduced or increased in size, while the spleen may be enlarged. Evidence of collateral circulation is seen in the enlargement of the mammary, epigastric, hypogastric and hemorrhoidal veins. Rarely is the superficial circulation so marked as to present the picture of a true caput medusae. Hematemesis or melena may occur, suggesting the presence of leaking or ruptured esophageal varices. As the condition progresses, the patient loses flesh and strength. The skin is muddy or icteroid in color, and the face is thin and shrunken, contrasting markedly with the full distended abdomen. The laboratory observations in a series of typical cases of this type are given in table 1. The clinical history is illustrated in case 1.

Case 1 (case 4, table 1).—A man, aged 63, had undergone herniotomy elsewhere in August, 1924, when free fluid was found in the abdominal cavity. Enlargement of the abdomen became noticeable soon afterward and increased progressively. Associated with the abdominal distention there was moderate edema of the feet and ankles, slight bloating and abdominal distress and loss in weight.

When the patient was first seen on Nov. 10, 1924, the emaciation of the face and arms contrasted markedly with the distention and enlargement of the abdomen. He was anemic, and the complexion was sallow though not distinctly jaundiced. Because of the marked ascites, none of the abdominal viscera could be felt. The superficial abdominal veins were dilated, and there was edema of the legs and lumbosacral region. The blood urea was 40 mg. and the serum bilirubin 1.4 mg. for each 100 cc. A "delayed direct" van den Bergh reaction was obtained. There was retention of phenoltetrachlorphthalein with a reading of 17 per cent.

Twelve liters of slightly turbid, straw-colored fluid was removed by paracentesis. The spleen could then be felt extending 8 cm. below the costal margin. The liver was not palpable. The fluid rapidly reaccumulated. Commencing November 20, treatment with merbaphen and ammonium chloride was instituted, a total of 12 cc. of merbaphen being given. Satisfactory diuresis followed, and on December 18 the patient weighed 150 pounds (68 Kg.), which indicated a loss of 45 pounds (20.4 Kg.). The abdominal distention had disappeared completely, and no fluid could be demonstrated. The spleen was enlarged, while the margin of the liver was palpable just at the costal margin. The evidences of collateral

4. Rolleston, H. D.: Diseases of the Liver, Gallbladder and Bile Ducts, ed. 2, New York, The Macmillan Company, 1914, p. 811.

TABLE 1.—*Portal Cirrhosis with Ascites (Small Liver)*

Case	Date	Age, Years	Sex	Edge of Liver Palpable, Cm.*	Ascites, Grade 0 to 4	Duration of Ascites, Months	Spleen Enlarged, Grade 0 to 4	Jaundice, Grade 0 to 4	Urine: Specific Gravity	Urine: Albumin	Urine: Bile Pigments	Blood Count: Hemoglobin, Per Cent	Blood Count: Erythrocytes, Millions	Blood Count: Leukocytes	Phenolsulphonphthalein, Per Cent	Blood Nitrogen Partition, Mg. for Each 100 Cc.: Nonprotein Nitrogen	Urea	Uric Acid	Creatinine	Amino-Acid Nitrogen	Bile Pigments (Serum): Serum Bilirubin, Mg. for Each 100 Cc.	Bile Pigments (Serum): Van den Bergh Direct Reaction†	Fructose Tolerance: Blood Sugar Mg. for Each 100 Cc.: Fasting	One Hour	Two Hours	Change	Phenoltetrachlorphthalein Dye in Serum, per Cent: Fifteen Minutes	One Hour	Two Hours	Dye in Urine, Mg.	Comment
1	7/ 2/24	62	M	0	3	2	2	0	1.019	1	+	65	3.18	7,000	..	30	43	3.2	2.0	5.6	**2.9**	+	90	115	106	25	18	**18**	12	**2.0**	Cirrhosis observed at operation
2	3/26/23	42	M	0	3	6	1	0	1.026	0	..	67	3.72	4,600	70	..	20	2.4	0.8	7.0	...	..	...	...	...	..	18	**18**	..	...	Necropsy; weight of liver, 997 Gm.
3	1/ 8/25	27	F	0	3	4	0	0	1.010	1	..	65	3.62	4,600	50	..	35	2.8	1.1	5.5	0.6	±	...	...	...	..	20	**17**	8	**1.8**	
4	11/21/24	63	M	0	3	5	2	0	1.025	1	..	55	3.46	3,000	45	38	41	2.5	1.4	...	1.4	±	...	...	...	..	17	**17**	11	0.9	Cirrhosis observed at operation
	12/ 1/24	..	..	+	1	..	2	0	1.014	0	..	60	4.00	6,600	..	23	34	2.9	1.5	5.6	1.6	±	89	**135**	102	**44**	14	**15**	10	0.3	Merbaphen 5 cc.
	12/18/24	..	..	+	0	..	2	0	1.018	1	..	60	3.28	6,600	..	65	42	...	1.3	...	1.8	±	...	...	...	..	17	**20**	10	0.0	Merbaphen 13 cc.; loss in weight 30 pounds
	4/ 2/25	..	..	+	1	..	2	0	1.027	0	0	58	2.74	3,300	..	..	34	...	...	...	0.9	±	...	...	...	..	12	**10**	3	0.3	Four months later
	7/ 9/25	..	..	+	0	..	2	0	1.018	0	..	40	2.14	3,500	45	..	71	4.0	1.7	...	0.7	0	...	...	...	..	8	5	2	...	Seven months later
	9/ 2/25	..	..	+	1	..	2	0	1.020	0	..	37	3.02	3,700	..	..	50	...	...	...	0.7	0	...	...	...	..	7	4	2	...	
5	9/ 2/25	36	M	0	3	3	1	0	1.020	0	..	50	3.47	5,000	45	45	18	...	...	...	**2.9**	+	...	...	...	..	12	**17**	9	...	Cirrhosis observed at operation
6	1/ 8/25	48	M	+	3	11	1	0	1.019	0	..	65	3.80	4,800	50	28	15	2.3	1.2	6.1	1.6	+	...	...	...	..	12	**13**	11	**1.1**	
7	4/ 3/23	60	M	0	2	1	1	0	1.013	4	..	76	4.58	9,600	50	..	30	...	...	6.9	...	..	...	...	...	..	..	**12**	..	...	Cirrhosis observed at operation
8	9/19/24	37	F	0	3	18	1	0	1.030	0	0	68	3.85	5,300	40	25	18	1.9	1.5	5.1	1.9	0	123	95	95	28	..	**8**	7	**1.8**	Cirrhosis observed at operation
9	2/15/24	68	M	0	3	4	0	0	1.025	1	..	72	4.26	5,300	35	41	21	4.4	1.4	...	1.5	0	145	**156**	104	11	9	**8**	5	0.5	Cirrhosis observed at operation
10	6/25/24	60	M	0	3	6	1	0	1.023	1	0	70	4.37	7,000	20	..	42	2.0	1.5	...	0.4	0	...	...	...	..	10	**6**	5	0.5	Cirrhosis observed at operation
11	3/10/24	42	F	+	3	4	1	0	1.023	1	..	70	4.80	11,600	40	29	24	...	...	...	1.2	0	110	...	...	..	6	3	4	**1.0**	Cirrhosis observed at operation
																															Portal Cirrhosis with Ascites and Jaundice
12	7/30/24	58	F	0	1	1	0	4	1.011	2	+	73	4.38	7,200	50	45	59	...	1.3	7.0	**32.8**	+	108	...	...	..	..	**35**	..	...	Necropsy; weight of liver, 817 Gm.
13	3/17/25	64	F	0	4	1	0	1	1.010	1	+	64	3.07	8,400	..	..	16	3.3	1.2	...	**4.6**	+	112	...	...	..	22	**28**	25	**1.2**	Necropsy; weight of liver, 850 Gm.
14	5/26/24	54	F	+	2	5	0	4	1.017	1	+	45	2.88	7,800	..	25	21	1.7	1.2	5.4	**13.6**	+	93	**147**	135	**54**	20	**20**	20	...	

* + indicates barely palpable.
† + indicates a "prompt" direct reaction; ±, "delayed" direct reaction, and 0, an indirect reaction.

circulation over the abdomen vanished. At this time the serum bilirubin was 1.8 mg. The phenoltetrachlorphthalein reading was 20 per cent.

The patient was dismissed in good condition. He continued to feel well and was strong and active. He returned on March 19, 1925, because of a slight gain in weight and abdominal enlargement. He weighed 10 pounds (4.5 Kg.) more than at the time of his dismissal, and a small amount of fluid could be demonstrated in the abdomen. His general appearance was much improved; otherwise examination showed his condition to be the same as when he was dismissed three months before. The serum bilirubin was 0.9 mg., with a delayed direct van den Bergh reaction. The dye retention was 10 per cent. Three injections of merbaphen produced marked diuresis and a total loss of 15 pounds (6.8 Kg.) in weight. The patient continued to improve following his dismissal until June 8, 1925, when he had a severe gastric or esophageal hemorrhage. He was weakened and confined to his bed for ten days at that time, but had been well thereafter. Because of weakness and a reduction of hemoglobin to 40 per cent, he was given a transfusion of 500 cc. of citrated blood on July 10.

On Sept. 2, 1925, the erythrocytes had increased to 3,020,000. The serum bilirubin was 0.7 mg. with an indirect van den Bergh reaction. The phenoltetrachlorphthalein reading was 4 per cent. Since then he has been able to carry on his usual activity, although progressive loss of strength has been associated with recurring gastro-intestinal hemorrhages, and the ascites has been more difficult to control.

This case has been of interest because of the prolonged period of observation and the unusually complete control of the ascites under treatment with merbaphen. It will be noted that there was no immediate change in the degree of dye retention during the period of removal of the ascitic fluid. This would seem to eliminate changes due to passive congestion of the liver such as were observed in case 17 (table 2). Subsequently the amount of dye retention has progressively decreased. This improvement in hepatic function indicated by the phenoltetrachlorphthalein test is important, for it suggests that there may have been regenerative changes in the liver during the period of observation. This case is also of interest in that alcohol could be excluded as a possible etiologic agent.

The laboratory observations in the other cases in this group were similar. The bromsulphthalein test was normal in one case, but in all the others this test showed a varying degree of retention of the dye. In two cases a slight increase in the serum bilirubin indicated the presence of latent icterus. A "prompt" or "delayed" direct van den Bergh reaction was obtained in the majority of instances even though the serum bilirubin was not increased.

PORTAL CIRRHOSIS WITH ASCITES AND LARGE LIVER

An increase in the size of the liver is readily distinguishable in many cases of portal cirrhosis. When abdominal distention is marked, it may be impossible to feel the liver, and it is only after relief from the ascites that the condition can be recognized. At present there is no other clear-cut distinction between this group and the preceding one, in which

the liver is reduced in size. The extent of parenchymal degeneration, the proliferative changes in the connective tissue, the contraction of scar tissue, the amount of hepatic regeneration and the presence or absence of fatty changes are the important factors which influence the size of the liver. For obvious reasons these factors will not be discussed in this communication.

The cases included in this group differ from those listed as of the Laennec type in that the edge of the liver was palpable at or several centimeters below the costal margin. The clinical histories are similar in the two series of cases. Enlargement of the liver from other causes, especially from cardiac failure with chronic passive congestion, must be excluded. Apart from the influence of such associated conditions, the size of the liver is of questionable value in the clinical classification of portal cirrhosis. The results of the different functional tests are shown in table 2. The clinical history is illustrated in cases 2 and 3.

CASE 2 (case 23, table 2).—A man, aged 38, came to the clinic on Oct. 18, 1920, because of a moderate loss of weight during the preceding year. He was constipated and complained of occasional "biliousness" and indefinite distress in the upper part of the abdomen. The edge of the liver was palpable at the costal margin. The patient was nervous and of introspective temperament. The diagnosis was indeterminate at that time. Six months later, pneumonia was followed by empyema which necessitated drainage. The patient was well for awhile, but in 1922 began to feel listless. His physician removed 6 liters of fluid from the abdomen at that time, and paracentesis was again carried out before readmission to the clinic on Oct. 1, 1923.

The abdomen was distended and tense, and the superficial abdominal vessels were dilated. After paracentesis and the removal of 3 liters of amber colored fluid, the edge of the liver could be felt 10 cm. below the costal margin, while the spleen was just palpable. The patient was not anemic. The specific gravity of the urine was 1.014, and only a trace of albumin was present. The blood urea was 43 and the fructose tolerance was normal. There was slight retention of dye, with a reading of 5 per cent.

Laparotomy was performed on Oct. 31, 1923, when 10 liters of ascitic fluid was removed. The liver was large and said by the surgeon to be cirrhotic. The spleen was about twice the normal size. It was removed and a Talma-Morison operation performed. Convalescence was uneventful. Fluid reaccumulated slowly at first, and paracentesis was necessary a month later. Thereafter recovery was uneventful. Three years later this patient reported himself in good health, free from ascites and again able to carry on his business.

CASE 3 (case 17, table 2).—A man, aged 63, first came to the clinic on Jan. 20, 1925, because of abdominal distention, which had come on two years before. Previously he had enjoyed good health, although he was a heavy user of alcohol. The onset of the illness was gradual, the distention first being noted in November, 1922. Subsequently paracentesis was carried out on eight occasions, from 8 to 10 liters of light-colored fluid being removed each time. The relief following such a tapping lasted for from five to six weeks. In August, 1924, a physician elsewhere administered calcium chloride as a diuretic. Seventy pounds (31.8 Kg.) were lost in four weeks, but the ascites returned when the calcium chloride was discontinued.

The patient's weight on admission was 250 pounds (113.4 Kg.), 60 pounds (27.2 Kg.) more than his best normal weight. There was marked ascites, and it was impossible to palpate any of the abdominal viscera. There was definite peripheral

arteriosclerosis, and the heart was somewhat enlarged. Considerable edema of the scrotum and legs was present.

There was moderate secondary anemia. An electrocardiogram showed auricular fibrillation, right ventricular preponderance and inversion of the T-wave in leads I and II. The specific gravity of the urine was 1.030, and a trace of albumin and casts were present. The blood urea was 54 mg. and the serum bilirubin 2.1 mg. A direct van den Bergh reaction was obtained. The retention of phenoltetrachlorphthalein was 25 per cent.

The patient was put on a low protein, salt-free diet, and the fluid intake was restricted. In addition, 10 Gm. of ammonium chloride was given daily, and merbaphen intravenously every fourth day. Improvement was rapid and striking. On February 13, after a total of 4 cc. of merbaphen had been taken, the patient's weight was 175 pounds (79.4 Kg.), a loss of 75 (34 Kg.) pounds. At this time the heart was regular and the edema of the extremities had disappeared, although digitalis had not been given or other specific cardiac medication instituted. Fluid could not be demonstrated in the abdomen. The edge of the liver could be felt 8 cm. below the costal margin, and the spleen was palpable 5 cm. below the left costal margin. The blood urea was 21 mg. and the serum bilirubin 1.6 mg., with a direct van den Bergh reaction. There was marked improvement in the phenoltetrachlorphthalein test with a reading of 10 per cent.

The patient continued to improve in strength and health while under observation. On March 3, the weight was 160 pounds (72.6 Kg.), indicating a total loss of 90 pounds (40.8 Kg.) during treatment. The serum bilirubin was 2.1 mg. and the dye retention 10 per cent. The patient was dismissed in good condition. He returned on June 23, 1926, for reexamination, having been in good health and free from ascites during the intervening period. The liver was reduced somewhat in size, extending only 2 cm. below the costal margin, and the spleen was no longer palpable. The blood urea was 31 mg. The serum bilirubin was 1.8 mg. and the van den Bergh reaction indirect. The phenoltetrachlorphthalein test gave a reading of 8 per cent.

When this patient was first seen, decompensation with passive congestion was superimposed on the cirrhosis. With the recovery of compensation and the disappearance of the hepatic congestion, the degree of dye retention decreased rapidly from 25 to 8 per cent. Thereafter the phenoltetrachlorphthalein test did not change. We believe that this sequence demonstrates both the temporary effect of passive congestion and the more permanent effect of cirrhosis on the excretion of dye. These observations are analogous to those of the phenolsulphonphthalein test in passive congestion superimposed on chronic nephritis.

These two cases serve to illustrate the functional changes observed in the nine cases included in this group. Retention of dye was uniformly present though to a varying degree. The serum bilirubin was occasionally increased to a degree indicating the presence of latent icterus, but for the most part it varied around the upper limits of the normal. A direct van den Bergh reaction was obtained in the majority of cases. The changes in the other laboratory tests were not striking, and will be discussed later.

PORTAL CIRRHOSIS WITH ASCITES AND JAUNDICE

Jaundice is an occasional but not a prominent symptom in portal cirrhosis. When "signal jaundice" is observed at the onset of the condition, it is usually of slight degree and transitory. During the progress of the malady the skin may acquire the muddy yellow or subicteric hue

that is so characteristic a feature of the hepatic facies. Manifest and outspoken jaundice, on the other hand, is uncommon and is usually considered a late or terminal event. Such cases, both clinically and from the point of view of the gross pathology, may present a picture distinguishable with difficulty from that seen in the end stages of biliary cirrhosis with terminal ascites. Differentiation must be made largely on the basis of the chronologic sequence of events in the history.

Case 4 (case 13, table 1).—A woman, aged 64, was admitted to the clinic on March 17, 1925, because of ascites and jaundice of two months' duration. In January, 1925, she first noted weakness and loss of strength, which increased progressively. In February, she began to feel that the abdomen was getting larger; there was also some swelling of the ankles. Shortly afterward jaundice appeared, but she did not have pain, fever or pruritus. The stools became lighter and the urine dark. The jaundice never became extreme and diminished slightly in intensity. The ascites increased, however, and strength failed rapidly.

The abdomen was greatly distended with marked ascites. The abdominal viscera could not be felt. There was edema of the lower abdominal wall and of the lower extremities. Moderate jaundice was evident. The urine contained bile and a trace of albumin. There was moderate secondary anemia. The serum bilirubin was 4.6 mg., and a direct van den Bergh reaction was obtained. There was retention of phenoltetrachlorphthalein with a reading of 28 per cent.

A Talma-Morison type of omentopexy was performed in an attempt to relieve the ascites. The surgeon reported the presence of advanced cirrhosis of the liver. The patient did not rally following the operation, but went into coma; the blood urea rapidly rose to 130 mg., and death occurred on the fourth day after operation. At necropsy the liver was found to weigh but 850 Gm.

The laboratory data in three cases are given in table 1. As would be expected from the presence of jaundice, the serum bilirubin was increased, and a direct van den Bergh reaction such as is encountered in obstructive jaundice was obtained. The retention of dye likewise was marked.

PORTAL CIRRHOSIS WITHOUT ASCITES

Obviously, the question of the early stage of portal cirrhosis has been omitted from consideration. In the absence of evidence of collateral circulation and ascites, it is difficult to be certain of the nature of the changes in an individual case. However, under certain conditions, particularly among heavy drinkers, the diagnosis at times may be made clinically with a reasonable degree of certainty. If the liver is enlarged, the possibility of chronic hepatitis or early cirrhosis must be considered in the diagnosis.

The common textbook picture of cirrhosis is that of the so-called "gin drinker's liver." While suggested by clinical observations, the presence of a direct etiologic relationship between cirrhosis and alcohol is by no means firmly established, and in several of the cases reported in this series we could not secure a history of alcoholism or of the use of alcohol in any form. On the other hand, we present here a case illustrative of a small group in which the associated enlargement of the

liver can best be explained on the basis of the history of prolonged and intensive abuse of the use of alcohol. It is impossible to distinguish between the relative rôle of hypertrophy and that of fatty infiltration in such enlargement without making a microscopic examination. Extended clinical observation, likewise, is necessary to prove that ascites is a later complication in such cases. Nevertheless, the implication remains that these cases represent an early stage of the group discussed as the hypertrophic form of portal cirrhosis without ascites. Our studies include seven cases in this group. The laboratory observations are shown in table 2, and the history of one case of this type is given. The cases are included as representing the early changes of portal cirrhosis. This we must admit is presumptive, and the true nature of the hepatic disease not known.

CASE 5 (case 26, table 2).—A man, aged 25, was admitted to the clinic on Jan. 15, 1924, because of discomfort of three months' duration in the right side of the upper part of the abdomen. He was the son of a saloon-keeper and was a "bootlegger" by occupation. He had been a steady drinker from the age of 10 and for several years had averaged one pint of whisky a day. Three months before admission, he began to have pain and discomfort along the right costal margin. This pain was continuous but never severe, except on two or three occasions when it became disabling. He vomited then and had diarrhea for a few days. He did not have jaundice at any time.

On examination the liver was found to extend 12 cm. below the costal margin. It was firm and not tender. The spleen was not palpable. Urinalysis showed only a trace of albumin. The blood count was normal. A fractional test meal showed absence of free hydrochloric acid. Fluoroscopic examination of the stomach was negative. The serum bilirubin was 0.7 mg. There was slight retention of phenoltetrachlorphthalein with a reading of 5 per cent.

While the clinical picture in this case is somewhat indefinite, the history of pronounced and persistent alcoholism and the enlargement of the liver makes the presence of cirrhosis probable.

NOTE.—The patient returned to the clinic in February, 1927. At that time there was marked ascites, and he presented a clinical appearance of a typical portal cirrhosis with ascites and an enlarged liver.

BILIARY CIRRHOSIS WITH EXTRAHEPATIC OBSTRUCTION

While ascites and the development of collateral circulatory channels are the chief signs of interference with the portal circulation and so of portal cirrhosis, jaundice is equally indicative of the disturbance in biliary secretion that accompanies biliary cirrhosis.

The best known type of chronic hepatitis with jaundice is the biliary cirrhosis produced by occlusion of the common bile duct. This has been produced experimentally by Wickham Legg,[5] Harley and Barratt,[6]

5. Legg, J. Wickham: On the Changes in the Liver Which Follow Ligature of the Bile Ducts, St. Barth. Hosp. Rep. **9**:161, 1873.

6. Harley, V., and Barratt, W.: The Experimental Production of Hepatic Cirrhosis, J. Path. & Bact. **7**:203, 1901.

TABLE 2.—*Portal Cirrhosis with Ascites (Large Liver)*

Case	Date	Age, Years	Sex	Edge of Liver Palpable, Cm.	Ascites, Grade 0 to 4	Duration of Ascites, Months	Spleen Enlarged, Grade 0 to 4	Jaundice, Grade 0 to 4	Urine: Specific Gravity	Urine: Albumin	Blood Count: Hemoglobin, Per Cent	Blood Count: Erythrocytes, Millions	Blood Count: Leukocytes	Phenolsulphonphthalein, Per Cent	Blood Nitrogen Partition, Mg. for Each 100 Cc.: Nonprotein Nitrogen	Urea	Uric Acid	Creatinine	Amino-Acid Nitrogen	Bile Pigments (Serum): Serum Bilirubin, Mg. for Each 100 Cc.	Van den Bergh Direct Reaction*	Fructose Tolerance; Blood Sugar Mg. for Each 100 Cc.: Normal	One Hour	Two Hours	Change	Phenoltetrachlorphthalein Dye in Serum, per Cent: Fifteen Minutes	One Hour	Two Hours	Dye in Urine, Mg.	Comment
15	3/24/24	33	F	4	3	2	1	0	1.024	1	88	5.56	9,600	80	29	11	1.9	1.4	6.3	1.8	0	...	...	...	..	23	25	20	...	Cirrhosis observed at operation
	4/10/24	..	..	3	3	..	1	0	1.018	1	87	5.55	7,400	..	26	10	2.0	1.2	6.3	4.3	0	90	122	119	32	..	19	15	3.8	
	4/28/25	..	..	10	0	..	2	0	1.016	1	58	3.75	8,600	50	40	32	2.4	1.0	6.4	4.7	±	...	...	...	..	25	28	17	3.1	
16	4/ 9/25	55	F	6	3	1	0	0	1.032	0	75	4.36	6,600	85	19	..	2.7	1.7	...	3.6	+	...	...	...	..	26	22	18	2.4	
17	1/21/25	62	M	8	4	24	1	0	1.030	0	70	3.84	5,200	..	54	54	3.8	2.2	5.9	2.1	+	...	...	...	..	22	25	22	0.5	
	2/13/25	..	..	8	1	..	1	0	1.022	0	70	3.63	9,200	40	..	21	...	...	...	1.6	+	...	...	...	..	10	10	15	0.3	Merbaphen 4 cc.; loss of weight, 70 lbs.
	3/ 3/25	..	..	8	0	..	1	0	1.022	0	..	....	.....	40	..	44	3.1	2.5	...	2.1	+	...	...	...	..	10	10	8	0.3	Merbaphen 6 cc.; loss of weight, 84 lbs.
	6/23/26	..	..	2	0	..	0	0	1.022	0	87	4.96	7,900	50	41	31	...	1.6	6.8	1.8	0	...	...	...	..	..	8	3	...	Fifteen months later
18	11/10/24	47	M	8	4	2	2	0	1.031	0	72	3.60	4,000	55	28	17	2.0	1.5	5.2	1.9	0	...	...	...	..	30	22	15	...	
	11/21/24	..	..	8	3	..	..	1	1.024	1	75	3.92	4,000	..	..	19	2.2	1.1	...	5.8	+	...	...	...	..	..	20	20	...	Merbaphen 2 cc.
19	4/28/25	55	M	3	4	4	0	±	1.020	0	58	2.49	3,600	65	..	28	2.6	1.0	...	3.3	±	...	...	...	..	14	17	16	2.2	
20	11/ 4/24	63	F	2	4	2	3	0	1.010	0	70	4.31	4,600	..	31	20	2.5	1.4	6.7	1.8	+	...	...	...	..	..	13	12	0.7	Cirrhosis observed at operation
21	5/12/24	48	M	4	3	2	0	0	1.030	1	70	4.16	5,700	..	26	31	1.6	1.4	...	2.3	0	...	...	...	..	13	13	9	1.4	
22	4/ 9/24	50	M	7	3	6	2	0	1.023	0	70	4.25	6,100	65	..	19	...	...	...	0.6	±	...	...	...	..	10	10	5	1.2	
23	10/ 1/23	41	M	10	3	12	2	0	1.014	1	85	4.75	5,000	..	..	43	4.6	1.7	...	...	..	103	117	100	14	10	5	0	0.1	Cirrhosis observed at operation
													Portal Cirrhosis Without Ascites																	
24	11/ 5/24	51	M	12	0	..	0	0	1.024	0	78	5.13	8,300	..	..	27	2.3	1.6	...	1.8	±	...	...	...	..	11	11	11	1.3	
25	10/16/23	53	M	4	0	..	2	0	1.008	1	52	4.10	6,700	40	..	..	...	...	...	...	..	106	115	93	9	9	5	0	0.0	
26	1/15/24	25	M	12	0	..	0	0	1.021	1	78	4.35	4,300	..	31	18	...	...	...	0.7	0	94	102	95	8	8	5	2	0.0	
27	1/16/25	54	M	5	0	..	0	0	1.021	0	72	4.13	7,300	..	..	25	4.8	1.5	5.9	0.2	0	...	...	...	..	9	5	2	...	
28	2/ 3/25	42	M	5	0	..	0	0	1.027	0	85	4.66	6,600	..	37	36	3.7	1.4	...	1.0	0	...	...	...	..	8	5	3	0.4	
29	3/16/25	40	M	15	0	..	1	0	1.010	2	51	3.68	12,500	..	..	32	4.1	1.3	...	0.1	0	...	...	...	..	7	3	0	0.5	Cirrhosis observed at operation
30	4/29/25	53	M	4	0	..	0	0	1.016	0	60	4.24	11,000	60	..	19	...	...	...	0.1	0	...	...	...	..	3	1	0	0.5	

* + indicates a "prompt" direct reaction; ±, "delayed" direct reaction, and 0, an indirect reaction

Charcot and Gombalt,[7] Ogata[8] and others. Mangelsdorff,[9] Ford,[10] Weber[11] and others have summarized the clinical observations on patients with complete obstruction of the extrahepatic bile ducts and, further, have emphasized the similarity to the changes observed in animals. Ford pointed out that this form of cirrhosis cannot be explained wholly by the damming back of bile, but that an added etiologic factor is present in the inflammation of the walls of the biliary channels. Originally reported as the result of complete biliary obstruction, the condition now is more frequently found in patients with recurring biliary colic and intermittent obstruction. W. J. Mayo[12] has especially emphasized the importance of considering biliary cirrhosis the end-result of infectious and obstructive processes secondary to disease of the gallbladder or common bile duct.

The clinical features of such cases are usually distinct. Most characteristic is the long-continued history of recurring attacks of temporary of incomplete biliary obstruction, usually from gallstones. Temporary jaundice commonly accompanies these attacks. Clinical evidence of infection with chills or so-called hepatic or Charcot fever is common. The length of the history and the failure of the jaundice to disappear completely between attacks speak for the presence of permanent hepatic damage.

CASE 6 (case 42, table 3).—*Chronic Cholangitis with Choledocholithiasis.* A man, aged 65, came to the Clinic on March 8, 1924, having suffered from chills and fever for six years. Ten years before, there were irregular attacks of bilateral pain in the upper part of the abdomen associated with nausea and vomiting. These occurred at intervals for two years, then ceased temporarily. Six years before admission he again began to have intermittent painless attacks beginning with a chill and followed by fever, profuse perspiration and prostration. Recovery usually took place within twenty-four hours. Jaundice occurred regularly during the attacks, icterus sometimes preceding the chill. Following this the urine would be dark for a day or so. The stools occasionally were light but never clay-colored. The jaundice was never extreme, but, on the other hand, never disappeared completely between attacks. The complexion was dark and the skin brownish yellow. The liver was slightly enlarged, and the edge was

7. Charcot and Gombalt: Note sur les alterations du foie consecutives a la ligature du canal cholédoque, Arch. de physiol. norm. et path. **18**:272, 1876.

8. Ogata, Tomosaburo: Beiträge zur experimentell erzeugten Lebercirrhose und zur Pathogenese des Ikterus mit spezieller Berücksichtigung der Gallenkapillaren bei der Unterbindung des Ductus Choledochus und der Ikterogenvergiftung, Beitr. z. Path. Anat. u. z. allg. Pathol. **55**:236, 1913.

9. Mangelsdorff, J.: Ueber biliäre Lebercirrhose, Deutsches Arch. f. klin. Med. **31**:522-603, 1882.

10. Ford, W. W.: Obstructive Biliary Cirrhosis, Am. J. M. Sc. **121**:60, 1901.

11. Weber, F. Parkes: On Biliary Cirrhosis of the Liver with and Without Cholelithiasis, Tr. Path. Soc. London **54**:103, 1903.

12. Mayo, W. J.: The Surgical Treatment of the Cirrhoses of the Liver and Their Complications, Ann. Surg. **68**:183, 1918; The Liver and Its Cirrhoses, J. A. M. A. **70**:1361 (May 11) 1918; Surgical Treatment of Hepatic Cirrhoses, Ann. Surg. **80**:419, 1924; The Liver and Its Cirrhoses, J. Iowa M. Soc. **16**:299, 1926.

rounded but not tender. The urine contained urobilin but no bile. There was moderate secondary anemia. The serum bilirubin was 1.8 mg. A direct van den Bergh reaction was obtained. The fructose tolerance was reduced, and there was retention of dye with a reading of 17 per cent.

At operation a large stone was found in the common bile duct together with a fistulous connection between the common bile duct and the duodenum. The stones were removed and the fistula closed. The patient did well for a time, but bronchopneumonia developed, and he died on the tenth day. At necropsy marked biliary cirrhosis was found. The liver weighed 1,442 Gm.

The laboratory observations in this case are characteristic of those in the thirteen cases comprising this group. Retention of bromsulphonphthalein was constant and marked. The serum bilirubin in general was slightly increased, though not to a proportionate extent. A reading of 10 mg. was seen in one case, and values within the normal limits were obtained in three. A direct van den Bergh reaction was obtained in all.

BILIARY CIRRHOSIS WITHOUT EXTRAHEPATIC OBSTRUCTION

Besides the obstructive type of biliary cirrhosis, there is another form in which there is no demonstrable obstruction of the extrahepatic bile ducts. There has been much uncertainty with regard to the diagnosis and classification of such cases. However, there unquestionably does exist a type of cirrhosis in which the liver is enlarged, firm in consistency, and in which no demonstrable obstruction of the hepatic bile ducts can be found at exploration or necropsy. The spleen is also enlarged, sometimes to a greater extent than is common in either the obstructive type of biliary cirrhosis or in portal cirrhosis. Chronic jaundice of variable degree persists throughout the course of the disease. In these cases, W. J. Mayo considers splenectomy of value if not too long postponed. He has also emphasized the frequency with which hemolytic jaundice and splenic anemia have been confused with this condition in the past, although the marked pruritus of the biliary cirrhosis should be of diagnostic significance. The melanoderma or pigmentation of the skin which is present, in addition to the staining with the bile pigments, is also characteristic.

This form of biliary cirrhosis is undoubtedly related to that described by Hanot.[13] The descriptions of the latter are not entirely clear, and there is not at present any sharply defined clinical and pathologic entity to which this name may be applied. A probable relationship to the cases of subacute hepatitis or intraphepatic jaundice, such as those reported from this service by Weir, must be admitted. It has seemed best, therefore, to avoid sharply defined differentiations or eponymic terms in discussing this group and to refer to the cases as examples of primary or nonobstructive biliary cirrhosis, as previously pointed out. Table 3 presents data relating to four such cases, one of which is presented in detail as follows:

13. Hanot, V.: La cirrhose hypertrophique avec ictére chronique, Paris, Rueff et Cie., 1892, p. 182.

CASE 7 (case 46, table 3).—A woman, aged 42, first came to the clinic on Aug. 22, 1924, because of abdominal distress of two years' duration. She had never been strong and had been troubled with nausea, anorexia, smothering sensations and other symptoms for many years. In 1920 eight teeth were removed, which necessitated six injections of serum for hemostasis. Three weeks later, the right antrum became infected; soon afterward generalized pruritus appeared and persisted for several months before icterus was noted. The jaundice was slight and variable in degree, but was associated with rather marked bronzing of the skin. In 1922 pressure in the right upper quadrant of the abdomen caused soreness; later there were occasional chills, which were accompanied by pain along the right costal margin and an intensification of the jaundice for several days. In January, 1923, one ovary was excised elsewhere; the gallbladder was reported normal and was not disturbed. The symptoms recurred rapidly, with bloating, epigastric soreness, slight jaundice and increased pigmentation of the skin. In March, 1924, a second operation was performed elsewhere, and the spleen was found to be enlarged. A section of the liver removed for microscopic examination showed biliary cirrhosis with diffuse infiltration of round cells, which was most intense in the periphery of the lobule.

The patient was an asthenic, undernourished woman with slight yellowish discoloration of the sclerotics and marked generalized brownish pigmentation of the skin. The color of the skin suggested addisonian pigmentation, although it did not have the characteristic distribution. The spleen was palpable 10 cm. below the costal margin. The liver extended 5 cm. below the costal margin and was smooth and firm, regular in outline, but slightly tender. Renal function was excellent. The serum bilirubin was 2.1 mg., and a direct van den Bergh reaction was obtained. There was retention of phenoltetrachlorphthalein with a reading of 20 per cent. Because of the evidence of hepatic damage found at the previous operation, further intervention was postponed, but the patient was kept under observation.

She returned on March 20, 1925, because of persistence of the pruritus, slight jaundice and abdominal pains. She was stronger than when first seen, but there had not been any change in her color. The liver was unchanged, but the spleen appeared to be slightly larger than before. The serum bilirubin was 1.7 mg., and there was dye retention of 13 per cent.

At operation, March 26, 1925, an enlarged, somewhat vascular liver was found. There were many adhesions from the former operation with thickening in the region of the common duct, but no stones could be felt. The spleen was large and soft, but was removed without difficulty. Microscopic examination of the spleen showed fibrosis with subacute splenitis.

The postoperative recovery was uneventful. The subicteric color of the skin disappeared in part, but the pruritus persisted. Two weeks after the operation, the serum bilirubin was 0.8 mg. Retention of dye, however, persisted with a reading of 19 per cent.

Seven series of tests were made in four cases. In all there was retention of dye. As in the cases of obstructive biliary cirrhosis the serum bilirubin was slightly, though not markedly, increased, and the van den Bergh reaction was direct.

BILIARY CIRRHOSIS WITH ASCITES

Although jaundice and ascites are present in this group, as in the group already discussed under portal cirrhosis, the relation of the two symptoms is somewhat different. In the present group jaundice has constituted a marked clinical feature of the disease, the ascites as a rule being of late occurrence.

TABLE 3.—*Biliary Cirrhosis with Extrahepatic Obstruction*

Case	Date	Age, Years	Sex	Edge of Liver Palpable, Cm.*	Jaundice, Grade 0 to 4	Intermittent Jaundice, Duration, Years	Spleen Enlarged, Grade 0 to 4	Ascites	Urine: Specific Gravity	Urine: Albumin	Urine: Bile Pigments	Blood Count: Hemoglobin, Per Cent	Blood Count: Erythrocytes, Millions	Blood Count: Leukocytes	Phenolsulphonphthalein, Per Cent	Blood Nitrogen Partition, Mg. for Each 100 Cc.: Nonprotein Nitrogen	Blood Nitrogen Partition, Mg. for Each 100 Cc.: Urea	Blood Nitrogen Partition, Mg. for Each 100 Cc.: Uric Acid	Blood Nitrogen Partition, Mg. for Each 100 Cc.: Creatinine	Blood Nitrogen Partition, Mg. for Each 100 Cc.: Amino-Acid Nitrogen	Bile Pigments (Serum): Serum Bilirubin, Mg. for Each 100 Cc.	Bile Pigments (Serum): Van den Bergh Direct Reaction†	Fructose Tolerance: Blood Sugar Mg. for Each 100 Cc.: Normal	One Hour	Two Hours	Change	Phenoltetrachlorphthalein: Dye in Serum, per Cent: Fifteen Minutes	One Hour	Two Hours	Dye in Urine, Mg.	Comment
31	7/ 5/24	50	F	+	1	5	0	0	1.025	1	—	60	3.66	5,600	..	34	39	3.8	1.9	5.8	**3.8**	+	...	...	...	..	36	**34**	34	**1.3**	Choledocholithiasis; operation
32	8/14/24	66	M	4	2	4	0	0	1.022	1	+	71	4.08	8,000	70	38	44	3.5	1.5	4.8	**5.7**	+	101	123	103	22	25	**33**	25	**4.2**	Choledocholithiasis; operation
	8/22/24	..	..	4	2	..	0	0	.....	..	..	..	.....	.....	..	40	47	4.6	2.0	4.9	**5.1**	+	...	...	...	..	..	**22**	20	**3.0**	
33	9/17/24	61	M	+	2	40	0	0	1.003	0	—	65	3.86	5,100	..	24	17	3.2	1.8	4.5	**5.5**	+	95	123	**137**	**42**	34	**24**	22	**1.8**	Choledocholithiasis; operation
34	2/25/25	47	F	4	2	10	1	0	1.024	0	+	76	4.14	6,900	45	..	28	2.8	1.6	...	**4.1**	+	112	...	...	..	22	**26**	26	...	Postoperative stricture; operation
35	6/ 9/24	49	F	2	±	1	0	0	1.020	0	0	77	4.00	6,700	..	24	27	2.5	1.5	5.0	**2.5**	+	86	**146**	100	**60**	25	**25**	17	**1.5**	Chronic cholecystitis; operation
36	2/14/25	62	M	10	±	10	1	0	1.026	0	..	76	4.54	4,800	60	..	12	3.0	1.1	...	**4.9**	+	...	...	...	..	15	**25**	..	**1.1**	Chronic cholecystitis; operation
37	3/ 4/25	56	F	+	±	15	0	0	1.022	0	..	75	4.06	6,300	..	..	16	3.5	1.4	...	1.4	+	...	...	...	..	20	**23**	20	**1.3**	Choledocholithiasis; operation
	3/20/25	..	..	+	0	..	..	..	.....	..	..	..	....	.....	..	..	27	2.5	1.3	...	1.0	+	...	...	...	..	10	**15**	13	**2.2**	Fifteen days after operation
38	6/17/23	62	F	+	±	10	1	0	1.016	1	..	61	3.90	17,200	60	...	19	1.6	1.5	5.5	...	..	...	...	...	..	20	**20**	20	0.3	Choledocholithiasis; necropsy
39	7/ 7/24	41	F	10	3	10	0	0	1.025	1	+	57	3.40	5,300	..	28	25	2.1	1.3	6.3	**10.4**	+	84	107	72	23	17	**20**	17	**7.5**	Postoperative stricture; operation

40	7/29/21	73	M	+	1	5	1	0	1.024	1	0	70	4.20	7,300	60	29	25	2.4	1.5	5.4	1.5	+	86	133	89	**47**	22	**17**	..	...	Chronic cholecystitis; necropsy
41	9/ 1/24	57	F	6	±	10	0	0	1.010	0	0	65	3.37	6,200	60	27	9	2.4	1.3	5.5	**2.6**	+	...	...	...	..	17	**17**	15	**1.8**	Choledocholithiasis; operation
42	4/15/24	65	M	+	±	6	0	0	1.020	1	0	63	3.52	8,500	..	27	24	2.9	1.4	5.9	1.8	+	109	**142**	115	33	10	**17**	11	0.6	Choledocholithiasis; necropsy
43	7/12/24	31	F	6	±	5	2	0	1.022	0	..	50	3.24	12,700	50	26	20	1.9	1.2	5.8	**2.6**	+	94	106	90	12	..	**12**	11	**5.4**	Chronic cholecystitis; operation
															Biliary Cirrhosis Without Extrahepatic Obstruction														
44	1/ 3/24	22	M	15	2	3	3	0	1.018	2	+	55	2.79	6,600	40	..	10	...	...	...	**7.6**	+	...	...	...	..	17	**25**	25	...	Cirrhosis observed at operation
45	10/21/24	60	M	2	±	0	0	0	1.011	0	..	72	4.01	7,600	..	25	29	3.0	1.6	6.5	**2.4**	+	...	...	...	..	14	**20**	15	**1.2**	Cirrhosis observed at operation
	10/27/24	..	..	2	±	..	0	0	1.008	0	..	..	....	.....	..	25	15	2.2	1.7	6.4	**3.9**	+	...	...	...	..	17	**23**	12	**1.8**	
46	8/30/24	42	F	5	2	2	1	0	1.012	1	—	72	4.20	5,300	75	..	32	3.8	1.3	...	**2.1**	+	...	...	...	..	..	**20**	20	...	Cirrhosis observed at operation
	3/19/25	..	..	5	1	..	2	0	1.026	1	+	70	3.71	5,700	40	..	43	4.5	1.2	...	1.7	+	81	...	...	..	12	**13**	11	**1.1**	
	4/ 8/25	..	..	2	0	..	..	0	.....	..	..	..	....	17,200	..	..	21	...	...	...	0.8	+	95	...	...	..	15	**16**	12	**1.9**	Two weeks after splenectomy
47	12/13/23	46	F	10	1	12	2	0	1.016	1	0	70	4.36	7,000	..	..	..	...	...	...	**2.1**	+	...	...	...	..	..	**18**	12	**3.4**	
															Biliary Cirrhosis with Ascites															
48	6/21/25	59	F	+	3	2	1	+	1.021	0	+	47	3.69	4,600	..	..	29	1.7	1.2	...	**12.5**	+	...	...	...	..	30	**27**	25	...	Ascites three weeks, obstructive type; chronic cholecystitis; operation
49	8/ 6/24	57	F	0	±	10	0	+	1.010	0	..	77	4.09	7,800	..	26	16	2.2	1.4	4.9	**4.3**	+	79	123	106	**44**	12	**17**	12	1.7	Choledocholithiasis; ascites, obstructive type observed at operation
50	12/ 3/24	53	M	8	1	8	3	+	1.026	0	..	59	3.65	4,600	..	..	25	3.3	1.5	...	**3.5**	+	...	...	...	..	12	**27**	22	**11.5**	Ascites three years, nonobstructive type

* + indicates barely palpable.

† + indicates a "prompt" direct reaction; ±, "delayed" direct reaction, and 0, an indirect reaction.

This combination of cirrhosis with icterus and ascites in each case represents, as a rule, the end stage with the maximum of hepatic damage. The clinical and pathologic pictures in these different conditions are usually characteristic, though mixed or intermediate forms are frequent. In this terminal period, however, distinctions disappear and differentiation must be made largely on the basis of the history.

CASE 8 (case 49, table 3).—*Biliary Cirrhosis with Extrahepatic Obstruction and Ascites.* A woman, aged 57, came to the Mayo Clinic on Aug. 6, 1924, because of attacks of biliary colic which had recurred for ten or twelve years. The attacks of severe epigastric pain with nausea and vomiting usually lasted two or three hours, and residual soreness persisted for several days. Eighteen months before admission, the attacks became more frequent. They were more severe, pain was referred to the right shoulder and back, and slight jaundice was occasionally observed afterward. Abdominal distention was noticed several months before admission.

The skin was yellowish and sallow, and there was definite icteric staining in the sclerotics. The abdomen was distended and contained a moderate amount of fluid. The liver and spleen were not palpable. A roentgenogram of the gallbladder area showed multiple stones. The urine was normal. The serum bilirubin was 4.3 mg. with a direct van den Bergh reaction. There was retention of phenoltetrachlorphthalein with a reading of 17 per cent.

A Talma-Morison type of omentopexy was performed on September 8. Several liters of ascitic fluid were removed at the time. The liver was small and was described by the surgeon as cirrhotic. The gallbladder contained stones. The patient did not gain strength following the operation. The ascites recurred, hydrothorax developed later and death occurred on September 12.

At necropsy, marked cirrhosis with ascites was found. The liver weighed 948 Gm. The gallbladder contained multiple stones. The appearance of the patient, the ascites and the small shrunken liver were almost similar to these observations in case 4 (case 13, table 1). The long history of attacks of gallstone colic, however, serves to classify this case as one in which biliary obstruction and infection predominated.

CASE 9 (case 50, table 3).—*Biliary Cirrhosis without Extrahepatic Obstruction but with Ascites.* A man, aged 51, first came to the Mayo Clinic in March, 1923, because of diarrhea and a mass in the upper left quadrant of the abdomen. He had had typhoid fever as a child. At the age of 25, he visited Cuba, where he was reported to have had malaria and yellow fever. In 1917 and at irregular periods thereafter, he suffered from severe diarrhea, with bloody mucoid stools. Examination in 1917 revealed slight jaundice and enlargement of the spleen. The gallbladder was drained surgically, and the jaundice disappeared temporarily; the diarrhea persisted.

When first examined the spleen was greatly enlarged and extended nearly to the umbilicus. The stools contained blood, and proctoscopic examination showed the presence of hemorrhagic proctitis. The urine was normal, and the patient was not anemic. Rectal irrigations and the administration of emetine caused partial relief from the intestinal condition.

The patient did not gain strength, however; periods of slight painless jaundice of several days' duration become progressively more frequent, and finally the jaundice became permanent, although it fluctuated in degree and was never marked. Ascites developed a month before his return on Dec. 3, 1924. At that time there was a brownish pigmentation of the skin in addition to slight icterus. The abdomen was distended with fluid, and the superficial veins were prominent. The spleen was greatly enlarged and extended 10 cm. below the costal margin. The edge of the liver was felt 8 cm. below the costal margin.

Bile pigments were demonstrated in both the urine and the stools. Moderate anemia secondary in type had developed; the erythrocytes numbered 3,650,000, the

leukocytes 4,600, and the hemoglobin was 59 per cent. The serum bilirubin varied from 2.5 to 5 mg. with a direct van den Bergh reaction. The retention of dye was 27 per cent.

The patient responded fairly well to treatment with merbaphen for a while, and the ascites partially disappeared. He returned home, but later began to fail, lost a little weight, became weak and drowsy and died in coma on Jan. 30, 1924. Necropsy was not performed.

Exact diagnosis in a case such as this is impossible without microscopic examination. The history of previous infection, the splenomegaly and the recurrent attacks of painless jaundice, which eventually became permanent, and the terminal coma permit one with assurance to make a clinical diagnosis of biliary cirrhosis of the nonobstructive type.

The ascites in the later stages is probably evidence of secondary interference with the portal circulation similar in origin to that seen in some cases of obstructive biliary cirrhosis. The development of marked ascites and an extensive collateral circulation within the abdomen likewise has been a feature of the terminal condition in dogs subsequent to ligation of the common bile duct.

COMMENT ON INDIVIDUAL TESTS

Fructose Tolerance.—In 1914, Rowntree, Marshall and Chesney [14] among others, pointed out the unsatisfactory character of the fructose tolerance test of Strauss in cases of cirrhosis. We have not found the study of the blood sugar curve according to the method of MacLean and de Wesselow [15] to increase the clinical value of the test. Ligation of the common duct in animals produces a progressive decrease in the fructose tolerance. In patients with obstructive jaundice, a greater number of positive tests were obtained than in normal persons, but the results were difficult to interpret. Similar results were obtained in the different varieties of cirrhosis. Two positive and two doubtful tests were obtained in a total of nine cases of portal cirrhosis, while two positive and three doubtful tests were obtained in a total of eight cases of biliary cirrhosis. As in the cases of obstructive jaundice, this proportion is not sufficiently great to make the test of diagnostic value in any individual case. There was apparently no significant disturbance of the fasting blood sugar level in any of the cases studied.

NITROGEN PARTITION OF THE BLOOD

Bollman, Mann and Magath [16] have reemphasized the importance of the liver in the formation of urea within the body. The blood urea

14. Rowntree, L. G.: Marshall, E. K., and Chesney, A. M.: Studies in Liver Function, Tr. A. Am. Phys. **29**:586, 1914.

15. MacLean, H., and Wesselow, O. L. V.: The Estimation of Sugar Tolerance, Quart. J. Med. **14**:103, 1920-1921.

16. Bollman, J. L.; Mann, F. C., and Magath, T. B.: Studies on the Physiology of the Liver: VIII. Effect of Total Removal of the Liver on the Formation of Urea, Am. J. Physiol. **69**:371, 1924.

level may be low in portal cirrhosis, in one case 9 mg., an observation in accord with the previous report of Rowntree, Marshall and Chesney. There is a decrease in the blood urea in dogs when the common bile duct is obstructed experimentally, and there is a tendency for the blood urea of patients with obstructive jaundice to fall within the lower limits of the normal. A similar change is observed in the present series of cases of biliary cirrhosis. The urea values observed in either type of cirrhosis are for the most part within the lower limits of normal. In view of the accompanying nutritional disturbance in so many of these cases, it is doubtful whether much diagnostic importance can be attached to these changes in the nitrogen partition of the blood. Furthermore, the terminal rise in blood urea observed in case 4 (case 13, table 1) is evidence that urea formation is by no means in complete abeyance. In consequence, we do not feel that changes in the nitrogen partition of the blood with a reduction in the proportion of urea are of great diagnostic import in the individual case.

The other nitrogenous constituents of the blood that were studied, namely, the total nonprotein nitrogen and the uric acid, creatinine and amino-acid nitrogen, did not show consistent variation from the normal.

Fibrinolytic Ferment.—Goodpasture [17] described the presence of a fibrinolytic ferment in the blood in cases of cirrhosis. Rowntree, Marshall and Chesney confirmed this observaion. We have not observed fibrinolysis in any other condition, and even in cirrhosis it is relatively infrequent. A positive test is perhaps diagnostic, but a negative test will not permit the exclusion of cirrhosis.

Serum Bilirubin.—The serum bilirubin was normal in amount in the majority of the cases of portal cirrhosis with ascites. The finding of a slight increase in the bilirubin in five instances, however, is in accord with the clinical observation that slight and transient jaundice not infrequently accompanies this form of cirrhosis. With the occurrence of frank jaundice in the terminal stages of this condition, the serum bilirubin becomes elevated (chart 1).

Biliary cirrhosis, whether of the obstructive or nonobstructive type, is usually accompanied by an increase in the serum bilirubin. This increase in general is moderate or slight, and comparable with the subicteric hue of the skin. The direct van den Bergh reaction was obtained in all the cases that showed an increase in the quantity of serum pigment. The degree of retention of pigment in these cases in general did not reach the high level seen in jaundice due to gross obstruction of the extrahepatic bile ducts. Moreover, it seemed to be largely

17. Goodpasture, E. W.: Fibrinolysis in Chronic Hepatic Insufficiency, Bull. Johns Hopkins Hosp. **25**:330, 1914.

independent of gross obstruction, since the stools were colored in the majority of these cases and bile-stained fluid could be obtained by duodenal drainage.

Phenoltetrachlorphthalein.—Rowntree, Hurwitz and Bloomfield [18] found that in hepatic cirrhosis there was a delay in the excretion of phenoltetrachlorphthalein. A retention of the dye in the blood serum has been pointed out by Rosenthal,[19] Ottenberg, Rosenfield and Goldsmith,[20] and others,[21] although the differentiation of various clinical types of cirrhosis has not always been sufficiently clear to permit the evaluation of the test in each (chart 2).

We have observed definite retention of dye in the cases of portal cirrhosis with ascites; but one case (case 11, table 1) was observed in which the reading was normal. No significant difference was observed between the observations in this group in which the liver was reduced and in the group in which the liver was increased in size. With the

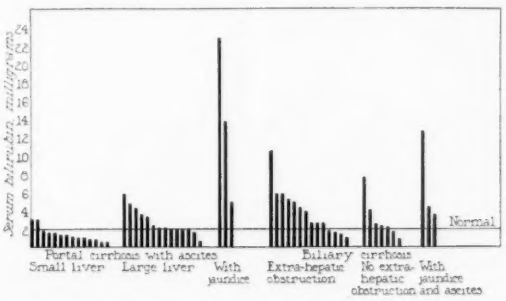

Chart 1.—Variations in the serum bilirubin in different types of hepatic cirrhosis; the maximum normal level is indicated.

development of jaundice there is an increase in the degree of retention of dye. This again is comparable with the results previously reported in obstructive jaundice. Only a slight degree of retention of dye was observed in the group of portal cirrhosis without ascites.

18. Rowntree, L. G.; Hurwitz, S. H., and Bloomfield, A. L.: An Experimental and Clinical Study of Phenoltetrachlorphthalein as a Test for Hepatic Function, Bull. Johns Hopkins Hosp. **24**:327, 1913.

19. Rosenthal, S. M.: A New Method of Testing Liver Function with Phenoltetrachlorphthalein, J. A. M. A. **79**:2151 (Dec. 23) 1922.

20. Ottenberg, R.; Rosenfeld, S., and Goldsmith, L.: The Clinical Value of the Serum Tetrachlorphenolphthalein Test for Liver Function, Arch. Int. Med. **34**:206 (Aug.) 1924.

21. Bull, D. C., and Bauman, L.: Results Obtained with the Phenoltetrachlorphthalein Test of Liver Function, Surg. Gynec. Obst. **40**:411 1925. Gilbert, A., and Coury, A.: L'exploration fonctionelle du foie par l'epreuve de la phénoltetrachlorephthaléine, Compt. rend. Soc. de biol. **90**:337, 1924. Gonzalez, A., and Karr, W. G.: A Comparative Study of the Phenoltetrachlorphthalein and Hemoclastic Tests of Liver Function, Arch. Int. Med. **34**:282 (Sept.) 1924.

It is difficult to explain fully the changes observed in the phenoltetrachlorphthalein test. The degree of retention of dye is greater in the presence of ascites. On the other hand, relief from the ascites did not affect the degree of retention in the majority of instances. The test in these cases is apparently independent both of the degree of interference with the portal circulation and of disturbances in the excretion of bile. It most closely corresponds to the extent of the degenerative processes in the liver, and, while it may be modified by regeneration, it is possible that the areas of regenerated hepatic cells are functionally less efficient than normal. This would explain in part the slight effects noted in relation to the size of the liver.

Following the experimental production of biliary obstruction, there is a close parallelism in the changes in the phenoltetrachlorphthalein test and the increase in the serum bilirubin. Similarly, there is a correspondence between the increase in the serum bilirubin and the degree

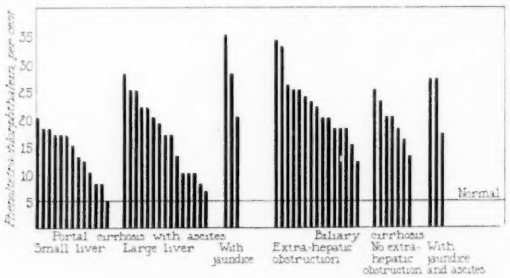

Chart 2.—Variations in the degree of phenoltetrachlorphthalein retention in different types of hepatic cirrhosis; the maximum normal level is indicated.

of retention of dye observed in cases of obstructive jaundice. In cases of biliary cirrhosis, retention of phenoltetrachlorphthalein uniformly occurred. It was more marked in this condition than in the cases of portal cirrhosis, an observation which may be related to the degree of retention of bile. At the same time we wish to point out that the degree of retention of dye is relatively greater than one would expect to find in a case of obstructive jaundice having the same degree of bilirubinemia. This disproportion between the serum birilubin and the phenoltetrachlorphthalein we believe to be evidence of the marked changes present in the liver. As such it may have clinical significance.

COMMENT

We have reported a series of cases of chronic hepatitis with inflammatory and proliferative changes of the type usually described under the head of cirrhosis. From the clinical point of view a detailed discussion

of the classification of such cases seems unwise at the present time, for intermediate and mixed forms occur, particularly in the terminal stages of the process; however, the presence or absence of ascites and jaundice and their sequence may serve for tentative division of cases into those of portal and biliary cirrhosis, respectively.

Functional tests may be taken as further evidence of the validity of this differentiation. In common with our experience in other hepatic conditions we have found it difficult to demonstrate consistently changes in the metabolic functions of the liver, with particular reference to changes in carbohydrate or protein metabolism. The most striking changes have been those shown by the use of phenoltetrachlorphthalein.

In advanced cirrhosis of either type, marked retention of dye occurred without reference to the presence of jaundice or ascites. In portal cirrhosis the striking functional observation is the high degree of retention in the absence of bilirubinemia. This evidence of dissociation of function is characteristic; similar changes have been observed previously by us in carcinomatous involvement of the liver without icterus. The degree of retention of dye in this series of cases of portal cirrhosis roughly corresponded to the degree of supposed hepatic damage, being slight in those cases without ascites which presumably were representative of an early stage of cirrhosis with minimal disturbance of hepatic function. At the same time, a direct relationship was not observed between the degree of retention of dye and the degree of ascites present. Furthermore, the complete disappearance of ascites is not accompanied by disappearance of the retention, and we feel that the circulatory disturbance responsible for the ascites is not of itself an index of the degree of functional damage to the liver. It is possible that the phenoltetrachlorphthalein test in cirrhosis is indicative of the functional hepatic damage and in a sense the balance between the degenerative and regenerative processes in the liver. It is of interest in this connection to note the decrease in the degree of retention of dye coincident with the clinical improvement in cases 1 (case 4, table 1) and 3 (case 17, table 2) in which medical treatment was instituted. At the same time, one must recognize that a definite relationship could not be found between the exact degree of retention of dye and the size of the liver.

In biliary cirrhosis, whether of the obstructive or nonobstructive type, the serum bilirubin was uniformly increased. This increase in general was slight, corresponding to the mild degree of chronic jaundice present in such cases. Extreme degrees of bilirubinemia such as occur in the so-called catarrhal jaundice or in some cases of malignant obstruction of the common duct were not seen. As in such cases, however, a direct van den Bergh reaction was present in all cases of biliary cirrhosis.

The phenoltetrachlorphthalein test is perhaps of less diagnostic significance in biliary cirrhosis because of the associated retention of bile. On the other hand, the degree of retention of dye is greater than ordinarily expected on the basis of the increase in the serum bilirubin. This disproportion apparently is related to the permanent changes in the liver and so may have some diagnostic value.

ANALOGY BETWEEN CERTAIN TYPES OF RENAL AND HEPATIC DISEASES

An analogy may well be drawn between abnormal conditions in the liver and those observed in the kidney. In cases of pyelitis or pyelonephritis there is an infection, which perhaps is primary in the pelvis, but which later spreads to the renal tissue proper. In consequence of the long continued infection and the intermittent obstruction, the parenchyma is destroyed and replaced by fibrous tissue, and a secondarily contracted kidney is ultimately produced. In chronic glomerulonephritis, on the other hand, there is evidence of repeated injury to the renal parenchyma by toxic or infectious agents. An inflammatory reaction with deposition of fibrous tissue follows, the epithelial degeneration leading in time to the formation of a contracted kidney. In arteriosclerotic or vascular disease of the kidney the interference with the renal circulation produces areas of malnutrition with consequent atrophy and scarring. This, too, leads to the development of a contracted kidney. In each instance laboratory evidence of functional disturbances indicates the severity of the process and confirms the clinical symptoms of renal insufficiency.

The gross appearance of some of these different types of contracted kidney may be similar, especially in the late stages. The functional changes are similar in all. If, as frequently occurs, a mixed or combined type of nephritis is present, it becomes difficult on pathologic evidence alone to determine the primary lesion, although this point may be clarified by clinical evidence, particularly concerning the chronologic sequence of events.

Obviously, care should be used in making an analogy between the diseases of these two organs and especially as regards the deductions drawn therefrom; nevertheless there are certain points of similarity. In this manner, it is evident that the pathologic processes responsible for obstructive biliary cirrhosis resemble those seen in chronic pyelitis or chronic pyelonephritis. The nonobstructive type of biliary cirrhosis and portal cirrhosis may be compared with chronic glomerulonephritis and the arteriosclerotic kidney, respectively. The cases of cirrhosis with terminal ascites and jaundice correspond roughly to the end-stage or contracted kidney. Combined or mixed forms of cirrhosis occur as do combined or mixed forms of nephritis.

In this terminal stage of cirrhosis, differentiation on clinical, functional or pathologic grounds may be impossible, and the nature of the initial process must be presumed largely from the history.

SUMMARY

We are impressed with the fact that the condition termed cirrhosis of the liver is an end-result of chronic inflammatory, degenerative and proliferative changes that may be produced by a variety of disease processes. The determination of the concentration of bilirubin in the serum and of the retention of phenoltetrachlorphthalein promises to be of assistance in the clinical study of such cases. The other tests of hepatic function studied have served to emphasize the importance of the great functional reserve of the liver. They failed, however, to show significant or sufficiently specific changes to make them of any great clinical value.

The phenoltetrachlorphthalein test is of particular value in the diagnosis and study of cases of portal cirrhosis with ascites. In such cases it is apparently an index to the existing functional balance between degenerative and reparative changes in the liver. As such, the observations are largely independent of the amount of ascitic fluid. One case is reported in which there was some evidence of improvement clinically and functionally and in which regenerative changes in the liver during treatment were assumed.

While jaundice is not a striking manifestation in portal cirrhosis, the study of the serum bilirubin showed the presence of latent icterus in several instances.

The study of the serum bilirubin has been of particular value in determining the degree of retention of bile in all types of biliary cirrhosis. Retention of dye is uniformly present but is more marked than would usually be expected from the degree of bilirubinemia. This disproportion is perhaps evidence of the pathologic changes in the liver.

In the end-stages of cirrhosis, irrespective of whether it is primarily portal or biliary in origin, the clinical picture and pathologic and functional changes may be well-nigh indistinguishable. Diagnostic differentiation is then based on the chronologic sequence of the more remote events as elicited in the history.

In cirrhosis of the liver functional studies are proving of considerable clinical importance, particularly from the standpoint of classification and differential diagnosis. They stimulate more serious clinical investigation and emphasize the need of focusing the attention of physicians on the problems of hepatic disease.

BLOOD SUGAR

COMPARISON OF BLOOD SUGAR CURVES FOLLOWING INGESTION AND INTRAVENOUS INJECTION OF GLUCOSE *

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The degree of hyperglycemia that follows the administration of glucose is commonly used as a measure of the efficiency of the sugar-utilizing mechanism of the body. The method ordinarily employed in performing this test is to measure the concentration of sugar in the blood at intervals after the ingestion of a certain amount of glucose. The criticism has long been made that the speed with which glucose is absorbed from the intestinal tract may vary. For example, Beeler and others [1] have demonstrated that an hour after the ingestion of glucose, from 22 to 68 per cent may be aspirated from the stomach. The amount may vary in different persons and in the same person from time to time. However, they fail to show that the amount of glucose recovered is inversely related to the height of glycemia. It is well known, on the contrary, that the ingestion of varying amounts of sugar above 50 Gm. or so makes little difference in the degree of the resulting hyperglycemia. Nevertheless, some workers prefer to administer the glucose by intravenous injection. Previous work on this subject has been adequately reviewed by Rigler and Ulrich,[2] and Jorgenson.[3] The essential difference between the two methods is that glucose given by mouth passes first through the liver, which it does not if introduced parenterally. We have seen no report concerning the use of both methods in the same subjects.

In the course of an investigation of the carbohydrate metabolism of persons subject to recurring convulsions,[4] we had occasion to employ

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* From the Laboratory of the Department of Neuropathology, Harvard Medical School and the Medical Service of the Massachusetts General Hospital.

* This research was made possible through a grant by the Committee on Epilepsy, New York City.

1. Beeler, C.; Bryan, A. W.; Cathcart, E. P., and Fitz, R.: An Improved Alimentary Glucose Tolerance Test, J. M. Research **1**:549, 1922.

2. Rigler, L. G., and Ulrich, H. L.: Blood Sugar Reaction Following Intravenous Injection of Glucose, Arch. Int. Med. **32**:343 (Sept.) 1923.

3. Jorgensen, S.: Comparison Between the Intravenous and Oral Application of Glucose for Loading of the Carbohydrate Metabolism, Acta Med. Scandinav. **55**:116, 1926.

4. Lennox, W. G.; O'Connor, M., and Bellinger, M.: Studies of the Metabolism in Epilepsy. III. The Blood Sugar Curve, Arch. Neurol. & Psychiat., to be published.

both methods in a large group of subjects. Because the comparative results obtained seemed to bear little relationship to epilepsy, they are presented here. This paper deals with 100 subjects whose blood sugar curves were drawn following both oral and intravenous administration of glucose. In all, 325 curves were made, 175 following ingestion and 150 following intravenous injection of glucose.

MATERIAL AND METHODS

Nine of the subjects were healthy men students. The rest, with a few exceptions, were patients with so-called epilepsy. None had diabetes. The tests were performed in the morning. Five cubic centimeters of venous blood were drawn from each subject when fasting, and at intervals of one-half, one and two hours after the administration of glucose. Specimens of urine were collected at the end of the two hour period. An amount of pure dextrose equal to 1.5 Gm. per kilogram of body weight in 33 per cent solution was ingested. For the injection tests, we used an amount of chemically pure anhydrous dextrose equal to 0.33 Gm. per kilogram of body weight in 20 per cent watery solution. In a few instances we buffered the glucose solution as recommended by Stoddard.[5] Because we feared that the injection of phosphate might modify results, and also because, in one instance, reaction occurred even with this solution, we discontinued its use. We followed the method of injection described by Rigler and Ulrich,[2] except that a syringe and threeway stop-cock were substituted for the pressure flask. Glucose was injected at the rate of 4 Gm. (20 cc. of solution) per minute. The average period of injection was approximately five minutes. In addition to the previously mentioned samples, blood was collected four minutes after the injection. Initial curves by the two methods were approximately equal in number. Reactions occurred in a little more than 5 per cent of the tests. On a given day, a reaction would occur in only one of several subjects, all of whom received the same material. Although many of the subjects received repeated injections, none had more than one reaction. The use of freshly distilled water, of old rubber tubing or of buffered solution did not prevent the appearance of an occasional reaction. The blood sugar curve of patients who had reactions was neither markedly abnormal nor markedly different from other curves made when no reaction occurred.

Blood sugar was measured by the method of Folin and Wu,[6] using the sugar tubes described by Evans,[7] and urine sugar by the method of Benedict, as recommended by Smith.[8] For the sake of brevity, we shall speak of blood sugar curves following ingestion and intravenous injection of glucose as ingestion curves and injection curves, respectively.

RESULTS OBTAINED

In table 1 we present intravenous curves of eleven healthy subjects, for nine of whom ingestion curves also were drawn. Two of us served as normal controls; the other subjects were medical students who had had recent physical examinations. So far as we are aware, there are no

5. Stoddard, J. L.: The Avoidance of Intravenous Glucose Reactions, Boston M. & S. J. **191**:1121 (Dec. 11) 1924.

6. Folin and Wu, H.: A System of Blood Analysis. A Simplified and Improved Method for Determination of Sugar, J. Biol. Chem. **41**:367, 1920.

7. Rothberg, V. E., and Evans, F. A.: A Modified Folin and Wu Blood Sugar Method, J. Biol. Chem. **53**:443 (Dec.) 1923.

8. Smith, M.: A Micro-Modification of the Method of Benedict for the Quantitative Determination of Reducing Sugar in Urine, J. Lab. & Clin. Med. **7**:3 (March) 1922.

published measurements of sugar curves from known normal subjects following intravenous injection of an amount of glucose similar to that used in these experiments. The seven nondiabetic patients whom Rigler and Ulrich [2] used were mostly subjects from whom abnormal reactions were to be anticipated (patients with hypertension, arthritis, cancer or of advanced age). The blood sugar had reached the fasting level in these patients by the end of two hours. Jorgensen and Plum [9] and Jorgensen [3] injected 20 Gm. of glucose dissolved in 50 cc. of water in their subjects and drew capillary blood for examination at frequent intervals. In their

TABLE 1.—*Blood Sugar Curves of Healthy Subjects Following Injection and Ingestion of Glucose*

		Blood Sugar, Mg. per 100 Cc. Blood								
			Minutes After Glucose Injected							
Subject	Amount Glucose Given, Gm.	Fast	4	15	30	60	90	120	Index Number	Urine Sugar, Gm.
1.....	23.3*	86	243	...	123	75	...	75	1.16	0.43
2.....	18.8	104	248	155	126	91	83	94	1.06	0.37
3.....	20.4	101	200	175	145	77	72	91	1.17	0.28
4.....	20.4	105	190	176	146	120	110	98	1.10	0.46
5.....	16.3	100	205	109	89	...	99	94	0.92	0.54
6.....	19.4	98	211	165	136	100	...	105	1.23	0.44
7.....	24.8	97	244	74	66	84	99	105	0.88	0.52
8.....	21.2	124	224	200	169	119	122	105	1.10	0.27
9.....	21.0	88	240	144	90	78	...	...	....	8.8
10.....	28.0	87	208	174	114	84	100	93	1.2	0.12
11.....	16.6	80	216	...	83	83	...	82	1.03	0.33
1.....	102.9†	103	...	...	167	116	...	83	1.21	0.15
2.....	84.8	116	...	126	132	143	...	133	1.19	0.13
3.....	91.8	112	...	...	133	164	...	140	1.36	0.30
4.....	91.8	110	...	...	133	119	110	100	1.06	0.13
5.....	81.6	99	...	108	133	78	87	80	1.08	0.05
6.....	97.2	102	...	130	165	116	108	102	1.31	0
7.....	111.6	101	...	133	100	100	76	109	1.08	0
8.....	95.3	128	...	213	239	132	143	138	1.47	0.14
9.....	46.2	105	...	116	102	133	85	...	....	0.24
Average injection...	20.6	100	223	149	121	93	95	96	1.08	1.34
Average ingestion...	89.2	108	...	138	145	122	111	111	1.22	0.13

* One-third gram per kilogram of glucose injected intravenously.

† One and five-tenths grams per kilogram of glucose ingested, except in subject 9 who received 0.75 Gm. per kilogram.

The measurements for subject 1 are the average of four intravenous and six ingestion curves, and for subject 2 the average of two intravenous and two ingestion curves.

The average values are for the first nine subjects. Missing 60 and 90 minute values are supplied by interpolation.

two published reports they tabulate observations on thirty-two nondiabetic patients, most of whom had no disease condition that might in itself give an abnormal blood sugar curve. In all but six of their patients, the blood sugar had returned to normal within ninety minutes.

Blood from our normal subjects was taken four, fifteen, thirty, sixty, ninety and 120 minutes after the injection of glucose. Blood sugar returned to the fasting level as follows: in fifteen minutes, one subject; in thirty minutes, three subjects; in one hour, six subjects and in two

9. Jorgensen, S., and Plum, T.: Differential Diagnosis Between Benign and Malignant Glycosuria by Means of Intravenous Injections of Small Quantities of Grape Sugar, Acta med. Scandinav. **58**:161 (May 22) 1923.

hours, one subject. Though our group of normal controls is small, we are forced to use it as a standard for judging the measurements from the larger group of patients. Chart 1 shows the average ingestion and injection curves of these nine subjects. It will be seen that although the glucose injected was but 24 per cent of the amount ingested, the peak of the average injection curve is much the higher. However, after the first precipitous fall of the injection curve, the two curves run a nearly parallel course.

Comparison of the injection and ingestion curves of each subject shows substantial agreement between the two methods. For example, both curves of subject 7 show that there was a remarkably quick disposal of sugar. Fifteen minutes after injection and thirty minutes after ingestion of glucose, the blood sugar was below the fasting level.

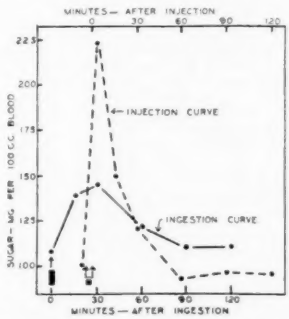

Chart 1.—The average blood sugar curves following the ingestion of 89 Gm. of glucose and the injection of 20 Gm. of glucose in the same nine healthy subjects (data from table 1). Ordinate represents milligrams of glucose per hundred cubic centimeters of whole blood. Abscissa represents minutes after administration of glucose. The two curves are placed so that their peaks correspond.

The data obtained from the whole number of subjects are too numerous to permit presentation in detail. For the purpose of comparison, we have endeavored to express the height of each blood sugar curve by a number. This figure, which we shall speak of as the index number, was obtained for ingestion experiments by adding the highest and the two hour measurements of blood sugar and dividing by two times the fasting value. In the case of intravenous tests, we feared that the sugar content of the blood drawn at four minutes might show variation because of incomplete mixing or because of the rapidly diminishing concentration of glucose, and used the half hour value as the highest. In several instances we were not able to calculate the index numbers of curves because two hour measurements were lacking. In charts 2 and 3 we have plotted the index numbers of the intravenous and the ingestion curves

against each other. In these charts, if the height of curves obtained by these two methods were proportional, the dots would fall in a zone extending from the lower left corner to the upper right corner of the chart.

Chart 2 presents the results obtained by plotting the index numbers of individual ingestion and injection curves against each other. Measurements of healthy subjects are represented by crosses, of patients on whom a single test by each method was made, by solid dots, and of those on whom repeated tests were made by circles. In the last group, the first injection curve of a subject was plotted against his first ingestion curve, the second injection curve against his second ingestion curve, etc. In seeking to obtain a standard of normal correlation of these index numbers, we drew a square in charts 2 and 3 which includes seven of the nine measurements from healthy persons listed

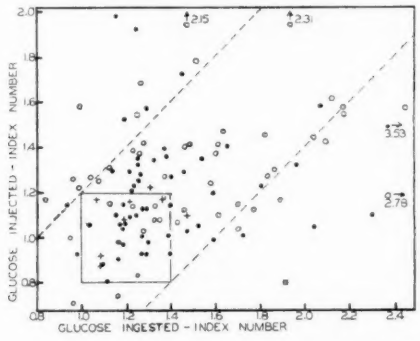

Chart 2.—Comparison of 115 measurements in which the height of ingestion and injection curves was compared. Ordinate represents the height of injection curves and abscissa the height of ingestion curves, as indicated by an index number. The index numbers were obtained by adding the highest value and the two hour value of curves and dividing by twice the fasting value. In the injection curves the half hour value was used in place of the highest. Crosses represent normal controls. Dots represents measurements of subjects on whom only one test was performed, whereas circles represent measurements of subjects on whom two or more tests by both methods were performed. The large square indicates limits of normal and the zone between broken lines the area in which there is correspondence between height of ingestion and injection curves.

in table 1. Of the remaining two, the ingestion curve of subject 9 was not strictly normal, and because the hypoglycemia that existed at the end of the first hour was not taken into account, the injection curve of subject 7 was really lower than the index number would indicate. The blood sugar curves represented by the limits of this square are within rather strict limits of normal. The parallel dotted lines extending from the corners of the square define a zone, values within which show some proportionality in the degree of hyperglycemia following ingestion and

injection of glucose. Inspection of chart 2 shows a wide scattering of the dots and circles. However, of the 115 measurements, eighty-six or 75 per cent are within the zone that we have marked as normal. Measurements of patients on whom repeated tests were made were more scattered than measurements of those on whom but a single test was made, probably owing to the fact that tests were repeated more frequently on patients who presented abnormal initial results.

As has been stated, most of these measurements were made on persons subject to convulsions. As noted in a previous paper,[4] the blood sugar curves of many of these patients presented abnormal variation in the level and configuration of successive curves. For this reason, it is probable that the measurements represented in chart 2 show a more widespread scattering than would measurements of patients whose physiologic functions were more stable. Some of this variability can be neutralized by taking the average of several curves rather than individual curves. This has been done in chart 3, which represents average

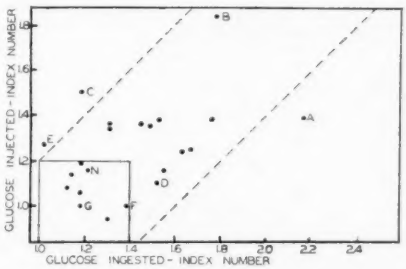

Chart 3.—Comparison of the average height of blood sugar curves (as represented by their index numbers) from twenty-two subjects on whom two or more tests by both methods were performed. The letters indicate the subjects whose blood sugar curves were presented in figure 4. *N* is a normal subject; the others are patients.

measurements of the twenty-two subjects on whom two or more blood sugar tests were made by each method. Of these twenty-two, nineteen or 87 per cent are within the normal zone. Therefore, in this group of nondiabetic patients, many of whom reacted to the administration of glucose in an abnormal manner, there is a fairly normal degree of proportionality in the height of blood sugar curves following ingestion and injection of glucose.

In order to vizualize more clearly the differences shown in some of these subjects, chart 4 presents the average curves of eight of the subjects whose measurements are on the periphery of the group shown in chart 3. The letters placed near the dots in chart 3 indicate subjects whose curves are represented in chart 4. The curves lettered *N* represent the average of six ingestion and four intravenous curves of one of

us. *A* represents the one extreme example of a patient whose average ingestion curve was disproportionally higher than her average injection curve. The average injection curve, judged by the curves of normal subjects in table 1, was slightly elevated, whereas her average ingestion curve was clearly diabetic in type. She showed no symptoms of diabetes except a gain of 28 pounds (12.7 Kg.) in weight during the twenty months of observation. In contrast with this is case *B*. Her average ingestion curve, though abnormally high, was lower than that of *A*, but during the first half hour following the injection of glucose there was distinct delay in the removal of glucose from the blood. Although this patient had no symptoms of diabetes, there was a family history of disturbance of the endocrine glands; namely, a sister had died of diabetes, her mother had exophthalmic goiter, and a brother had myxedema. Patients *C* and *E* had low ingestion curves and moderately

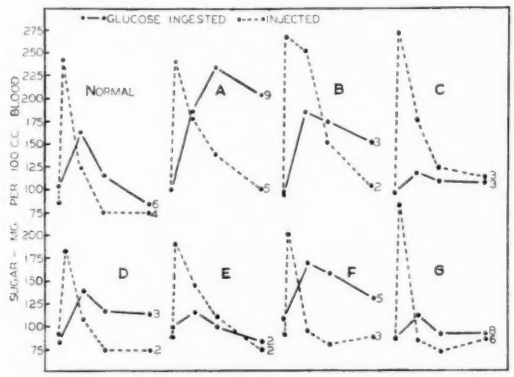

Chart 4.—Average ingestion and injection blood sugar curves of eight subjects, the index numbers of whose curves are compared in chart 3. The numbers accompanying each curve indicate the number of tests which were performed by each method. The curves drawn are the average of these separate curves.

elevated injection curves. Case *G* had curves which were low and exactly proportional. Regarding the several subjects in whom there was a discrepancy between the height of ingestion curves and of injection curves, we do not know the cause of this discrepancy or which of the two methods gages the efficiency of the sugar regulating mechanism more correctly.

Chart 5 is given as an example of the fact that both ingestion and injection curves may give intermittent evidence of delay in the removal of glucose from the blood. The figure records five ingestion and four injection curves of a patient who had essential hypertension and epilepsy. In the first two ingestion curves, numbered *1* and *5*, the concentration of blood sugar at one hour was much greater than at one-half hour.

Two of the injection curves also, numbered 3 and 4, showed similar delay. For thirty minutes after the injection, the concentration of glucose in the blood remained stationary. Five other curves of this subject were not markedly abnormal.

The question next arises concerning the degree of dispersion of values obtained in a group of subjects tested by the two methods. Jorgensen [3] has compared his curves drawn for nondiabetic patients following intravenous injection of 20 Gm. of glucose with Hagedorn's [10] curves from normal subjects following ingestion of 1 Gm. of glucose per kilogram of body weight. Jorgensen's curves show 20 per cent deviation from the mean against a 39 per cent deviation for those of Hagedorn. This comparison is open to criticism because one observer used patients and the other used normal subjects. Our observations are free from this objection because the same subjects were used for both tests.

Chart 6 represents the comparative frequency of ingestion and injection curves of various heights in our group of 100 subjects. Index

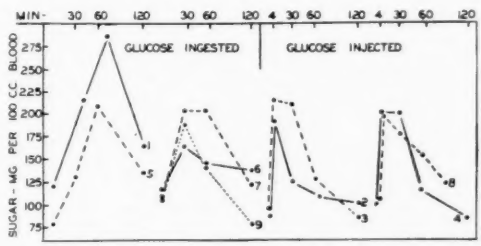

Chart 5.—Five injection and four ingestion curves of a patient with essential hypertension and epilepsy made during a period of twenty months. Certain curves by each method show initial delay in the removal of sugar from the blood. The numbers indicate the order in which the tests were performed.

numbers of 122 injection and 154 ingestion curves are plotted. Because our method of computing index numbers is arbitrary, the two curves are placed so that their apexes coincide. It will be seen that the distribution of normal curves (those with index numbers of 1.6 or below) is much alike for the two methods. However, the proportion of curves with index numbers above 1.6 is 18.5 per cent for ingestion and 5.6 per cent for injection curves. This indicates that the ingestion method gives a greater proportion of higher curves. We have pointed out elsewhere [11] that the difference between successive curves was less for injection than for ingestion curves. Injection tests did not give so many curves that were high only on initial trial. This smaller degree of variation in results

10. Hagedorn, H. C.: Undersøgelser vedrørende blodsuhkerregulationen hos menneshet, Thesis, Copenhagen, 1921.

11. Lennox, W. G., and Bellinger, M.: Repeated Blood Sugar Curves in Nondiabetic Subjects, to be published.

obtained by intravenous injection of glucose may be due, of course, to the much smaller amount of glucose used in the intravenous tests. In another article [12] we made a comparison of duplicate curves following ingestion and injection of the same amounts of glucose.

RENAL THRESHOLD

The older literature dealing with glycosuria following intravenous injections of glucose has been reviewed by Allen [13] and Kleiner.[14] Various authors have suggested that renal permeability may be altered for glucose which is introduced parenterally. In 1913 Thannhanser and Pfister [15] observed that when glucose was injected intravenously, the assimilation limit was only about 20 Gm. More recently Rosenberg [16] and Varela and Rubino [17] have shown that although glucose given by rectum may result in only a slight increase in blood sugar, glycosuria may occur. We have seen no observations in which results following

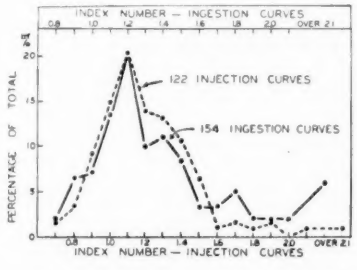

Chart 6.—Comparative frequency of various heights of 122 injection and 154 ingestion blood sugar curves. Ordinate represents the percentage of the total number of curves. Abscissa indicates the index numbers of curves. The scales are arranged so that the apexes of the two curves coincide. The two curves are roughly parallel, except that there is an undue proportion of high ingestion curves.

both enteral and parenteral administration of glucose were secured from the same group of subjects.

Table 2 presents the observations regarding the presence or absence of glycosuria in 160 ingestion and 137 injection experiments. There is

12. Lennox, W. G., and Bellinger, M.: Stimulation of the Sugar Regulating Mechanism as Shown by Duplicate Blood Sugar Curves, to be published.

13. Allen, F. M.: Glycosuria and Diabetes, Harvard University Press, 1913.

14. Kleiner, J. S.: The Disappearance of Dextrose from the Blood After Intravenous Injection, J. Exper. Med. **23**:509, 1916.

15. Thannhanser, S. J., and Pfitzer, H.: Ueber experimentelle Hyperglykämie beim menschen durch intravenose Zuckerinjektion, München. med. Wchnschr. **9**:2155, 1913.

16. Rosenberg, Max: Ueber die praktische Bedeutung der allmentaren Hyperglykämie Kurve, Klin. Wchnschr. **1**:360 (Feb. 18) 1922.

17. Varela and Rubino: Rektale Dextrosezufuhr und Blutzucker, Med. Klin. **18**:831 (June 25) 1922.

evident contrast between the incidence and degree of glycosuria in the two groups. Thirty-four per cent of the ingestion experiments were associated with glycosuria, against 85 per cent of the injection experiments. Comparison of the total results is hardly fair, however, because concentration of blood sugar in most of the injection curves was above, and in ingestion curves below, 200 mg. However, there is marked contrast in the proportion of subjects showing glycosuria at various levels of blood sugar. In the ingestion tests, the proportion presenting glycosuria decreased as the level of the blood sugar fell. For example, with blood sugar above 200 mg., 59 per cent of these subjects had glycosuria, whereas with the blood sugar below 200 mg., only 30 per cent had glycosuria. In the injection experiments, on the other hand, the elevation of the blood sugar made little difference in the incidence of glycosuria. Above 200 mg. the proportion was 87 per cent, below 200 mg., 83 per

TABLE 2.—*Proportion of Subjects Showing Glycosuria Following Ingestion and Injection of Glucose*

Highest Blood Sugar, Mg.	Glucose Ingested					Glucose Injected				
	Total Measurements	No Sugar	Sugar Present			Total Measurements	No Sugar	Sugar Present		
			Number	Per Cent	Average Amount, Gm.			Number	Per Cent	Average Amount, Gm.
More than 250	6	2	4	66	1.26	42	9	33	78	1.31
201 to 250	16	7	9	56	0.39	72	6	66	92	1.10
176 to 200	20	12	8	40	1.27	17	3	14	82	0.80
151 to 175	33	24	9	27	0.25	6	1	5	83	0.83
Less than 150	85	60	25	29	0.28					
Total	160	105	55	34	0.53	137	19	118	85	1.12

cent. Though the amount of glucose excreted was small in both groups, the average amount was twice as much for injection as for ingestion tests; namely, 1.12 Gm. for the former against 0.53 Gm. for the latter.

These differences are more significant when one remembers that in each test nearly five times as much sugar was ingested as was injected, and in the ingestion experiments the high levels reached were maintained for a longer time than in the injection experiments. Again, though in our injection experiments we always obtained blood at or near the peak of the curves, in our ingestion experiments hyperglycemia may have been greater than our measurements, made at half hourly intervals, would indicate.

The more ready excretion of intravenously injected glucose may possibly be explained by the fact that the rate of injection (4 Gm. a minute) presumably was faster than the rate of absorption from the intestines. That a rapid rate of introduction of glucose into the circulation is not necessarily associated with increased glycosuria, is suggested by the experiments presented in table 3. In subject *A*, 3 Gm. of glucose per kilogram (186 Gm. in all) were injected at the rate of 6 Gm. per

minute, and 11.4 Gm. of glucose were excreted. In subject *B* an equal amount of glucose per kilogram (205 Gm. in all) was injected at half this rate (or 3 Gm. per minute), and 15.1 Gm. were excreted. In these experiments the glucose was given in two equal doses. The interval between injections was approximately an hour. In each case glycosuria was greater with the second injection. This is seen more clearly with subject *B*. Although hyperglycemia was not nearly so high after the second injection of glucose, the amount of sugar excreted was twice as great. These two subjects showed little or no glycosuria when an equal amount of glucose (3 Gm. per kilogram) was ingested. These observations, though few, suggest that the slower entrance of glucose into the blood stream may not be entirely responsible for the smaller degree of glycosuria which accompanies the ingestion of glucose.

TABLE 3.—*Glycosuria with Reference to Duplicate Injections of Large Amounts of Glucose at Different Rates*

Subject	Rate and Period of Injection of Glucose	Period of Collection of Urine, Minutes	Limits of Blood Sugar, Mg.	Glucose in Urine, Gm.
A	6 Gm. per minute for 15 minutes	50	606-150	4.86
	6 Gm. per minute for 15 minutes	50	574- 95	5.71
		50	95- 48	0.84
Total	186 Gm. (3 Gm. per kilogram)	150	606- 48	11.41
B	3 Gm. per minute for 35 minutes	45	505-275	4.41
		45	275- 71	0.62
	3 Gm. per minute for 35 minutes	45	358-215	9.69
		45	215- 63	0.38
		10	65- 50	0.0
Total	205 Gm. (3 Gm. per kilogram)	190	505- 50	15.10

In the injection experiments summarized in table 2, glucose was injected at the same rate in all; yet there was lack of correlation between the incidence of glycosuria and the elevation of blood sugar. It is possible, as various writers have suggested, that glucose which first passes through the intestines and liver is modified in such a manner that it is less readily excreted than glucose which is introduced directly into the blood stream. In view of these observations, the assumption of Woodyatt and others [18] that the ability of the body to assimilate ingested glucose can be measured by the appearance of glycosuria following its injection, is permissible.

COMMENT

Because none of our subjects had diabetes, the data here presented cover rather a narrow range with regard to the degree of abnormality of the blood sugar curves. So far as we could judge from the use of

18. Woodyatt, R. T.; Sansum, W. D., and Wilder, R. M.: Prolonged and Accurately Timed Intravenous Injections of Sugar, J. A. M. A. **115**:2067 (Dec. 11) 1915.

the two methods in this group of nondiabetic subjects there was a fairly close agreement between the two in most instances. At any rate, the discrepancies between the two methods were no greater than the variations between successive tests by the same method.

The amounts of glucose that we injected were proportional to the weight, and therefore to the blood volume, of the subjects. If all the glucose were to remain in the blood until four minutes after the injection, the concentration of sugar in the blood would be increased approximately 350 mg. per hundred cubic centimeters. For example, the average amount of blood sugar in nine normal subjects would be 450 mg. after the injection; instead, it was 223 mg., as shown at the bottom of table 1. Therefore, during the nine minutes which elapsed after the injection was begun approximately one half of the injected glucose had disappeared from the blood. The rate of disappearance during this period varied in different subjects, as may be seen by a comparison of the four minute values in table 2 and in chart 4.

With regard to the comparative usefulness of the two methods, we may summarize the advantages of the intravenous route as follows: This method offers accurate dosage. The injection of 20 Gm. of glucose results in as high a blood sugar curve as the ingestion of 100 Gm. (chart 1); therefore, the intravenous route is particularly useful when, as in testing diabetic patients, one wishes to administer a minimum amount of glucose. When blood is taken at frequent intervals during the course of the tests, the intravenous curve is much smoother than the ingestion curve. Intravenous curves exhibit less variation from subject to subject, and in the same subject less variation from time to time.

As opposed to these advantages, the intravenous method has the following disadvantages: It requires more time and trouble of the investigator, and subjects those being investigated to an occasional reaction. More serious is the objection that glucose which is injected directly into the blood stream is in the nature of a foreign body. When glucose is injected, glycosuria is more pronounced and not so dependent on renal threshold. Also, as we suggest elsewhere,[12] injected glucose may have a less stimulating effect on carbohydrate metabolism than ingested glucose. These observations suggest that glucose which enters the body by way of the intestines and passes first through the liver is treated in a somewhat different manner from glucose which is introduced directly into the blood stream. A blood sugar curve test following intravenous injection of glucose would, therefore, seem to be less nearly physiologic than one following its ingestion. This consideration is largely theoretical.

If, as our study suggests, there is general correspondence in results obtained by both methods, choice of method is largely a matter of convenience rather than of principle. We believe that from the standpoint

of diagnosis, liberal interpretation of blood sugar curves and repetition of abnormal initial curves is of more importance than the method used.

SUMMARY AND CONCLUSIONS

1. We have plotted the blood sugar curves of 100 nondiabetic subjects following both intravenous injection and oral administration of glucose.

2. Of 115 curves, drawn by both methods, which could be plotted against each other, there was a normal degree of correlation in approximately 75 per cent. The average curves of twenty-two subjects on whom repeated tests were made by both methods showed normal correlation in 87 per cent.

3. When glucose was injected, glycosuria was more frequent and more pronounced than when glucose was ingested, and bore little relationship to the concentration of sugar in the blood.

4. The comparative advantage of the two methods are discussed.

BLOOD BILIRUBIN

ESTIMATION AND CLINICAL SIGNIFICANCE *

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Bilirubin is the most important of the bile pigments, the remaining two, biliverdin and bilicyanin, being oxidation products of bilirubin. It occurs in the blood as an acid and is therefore capable of forming salts differing in properties from the free acid. It is produced as the result of the break-down of the red cells of the blood; this probably occurs in the ramifications of the reticulo-endothelial system and has been shown to be almost entirely extrahepatic.[1] The bilirubin so produced is carried in the blood stream as a colloidal suspension and excreted from the liver into the bile passages. Before this excretion occurs, it undergoes a transformation in the liver cells from the form in which it is found in the blood to that in which it appears in the bile. The exact nature of this conversion is a matter of dispute, but the recent work of Collinson and Fowweather [2] tends to show that it consists of a change of the acid bilirubin into a soluble alkaline salt by union with ammonia. As they point out, it has been demonstrated in connection with dyes used for visualization of the gallbladder that only those which form true solutions are excreted with any degree of rapidity. They advance considerable evidence in support of their theory, which apparently warrants serious consideration.

The quantitative determination of blood bilirubin as a routine laboratory measure is a comparatively recent development. It is based on the van den Bergh reaction for the estimation and recognition of bilirubin in blood serum.[3] This estimation, on which the majority of figures for blood bilirubin previously noted have been based, was later pointed out by van den Bergh to be far from accurate; frequently as the result of a loss of bilirubin in the albuminous precipitate formed. In cases of a direct or biphasic reaction, this loss is considerable, although not so marked when the reaction is indirect. The technic of Thannhauser and Andersen [4] has overcome this objection by first mixing the diazo reagent with the test plasma, then adding the alcohol and saturated

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*From the Department of Internal Medicine, University of Michigan, and Clinical Laboratories, University Hospital.

1. Mann, F. C.; Bollman, J. L., and Magath, T. B.: Am. J. Physiol. **68**:114, 1924.
2. Collinson, G. A., and Fowweather, F. S.: Brit. M. J. **1**:1081, 1926.
3. Van den Bergh and Snapper: Deutsches Arch. f. klin. Med. **110**:540, 1913.
4. Thannhauser, S. J., and Andersen, E.: Deutsches Arch. f. klin. Med. **137**:179, 1921.

ammonium sulphate. After shaking and centrifugalizing, the clear alcoholic solution of azobilirubin is on the surface with the ammonium sulphate at the bottom and an intervening layer of white albuminous precipitate. McNee [5] found that this estimation gave results varying from as high as three times that of the original van den Bergh method in blood showing a direct or biphasic reaction, and 25 per cent higher in blood giving indirect reactions. Similar results were obtained in our laboratory.

A study was undertaken of bilirubin levels in the normal person and in a series of cases in which abnormal results were to be anticipated. The method used was essentially that of Thannhauser and Andersen, with some modifications.

METHOD

To 2 cc. of oxalated plasma is added 2 cc. of diazo reagent, freshly prepared. After from two to three minutes 5 cc. of 95 per cent alcohol and 2 cc. of saturated solution of ammonium sulphate are added. The solution is centrifugalized after shaking. The supernatant fluid is carefully drawn off and compared in a colorimeter, preferably of the Du Bosq type as more accurate, or in a Hellige colorimeter, with the standard solution described in the following. The result is multiplied by 4, none of the azobilirubin being dissolved in the ammonium sulphate, and expressed in mg. per thousand cubic centimeters of blood.

The use of plasma instead of serum obviates to a great extent the occurrence of hemolysis which interferes with the reaction and also gives a cloudy supernatant fluid. Even with the use of oxalated blood, early centrifugalizing is advisable, although not absolutely necessary.

The standard is made by dissolving 2.161 Gm. of anhydrous cobalt sulphate in 100 cc. of distilled water. The salt must be absolutely anhydrous and chemically pure, and when possible the solution should be checked against pure bilirubin. This standard gives a color equivalent to that given by 5 mg. of bilirubin per thousand cubic centimeters of blood, which constituted the old van den Bergh "unit." Various strengths of the aforementioned standard may be prepared for convenience in reading higher values of the unknown. When dilution of the unknown is necessary, 65 per cent alcohol should be used.

This standard is permanent if kept in the dark. The advantages over the ethereal solution of iron rhodanate originally used are obvious as regards both accuracy and convenience.

The use of this method does not interfere with the determination of the qualitative van den Bergh reaction. If on addition of the diazo reagent to the plasma, a reddish violet color appears within thirty seconds, the reaction is direct. If the color appears later than this and continues to deepen, with a further increase on addition of the alcohol, the reaction should be classed as biphasic. If a color does not develop on addition of the diazo reagent, the reaction is, of course, indirect.

The direct reaction is given by bilirubin only in the form in which it appears in the bile, that is, in cases of obstructive jaundice. There is

5. McNee, J. W., and Keefer, C. S.: Brit. M. J. **2**:52, 1925.

considerable evidence for the assumption that the biphasic reaction seen in arsenical jaundice, catarrhal jaundice and other cases of toxic or infective liver disease is the result of damage to the excreting cells of the liver, so-called hepatitis. The interpretation of the biphasic reaction, however, must be absolutely clear to avoid error, as reactions simulating the biphasic are given in cases of obstructive jaundice in which the patients are recovering. In cases in which an indirect reaction alone is obtained, all the bilirubin present is the result of hemolysis, in a normal or abnormal degree.

NORMAL VALUES

With the object of definitely determining the normal range using this method, the blood of fifty presumably normal persons, medical students, interns and others was examined. With one exception, the highest reading was 3.5 mg., the lowest 0.5 mg. Eight showed less than 1 mg., twenty-nine between 1 and 2 mg. and twelve 2 mg. and over. The one exception had a bilirubin of 7.5 and 8.5 mg. on two occasions some weeks apart. He was born in India, and he had never been found to have jaundice on clinical examination. The blood counts, smear and other laboratory tests did not show anything of note; the fragility test showed hemolysis commencing at 0.44 per cent and complete at 0.32 per cent. The liver was somewhat enlarged, and the case was probably one of chronic hepatitis.

Race and sex did not have any apparent influence, four negroes, two Orientals, and representatives of various other races being included in the series. The influence of pigmentation was not so marked as I had been led to expect, but definite brunettes tended to give slightly higher values. Blonds averaged 1.2 mg., the intermediate group 1.3 mg. and brunettes 1.8 mg.

Repeated readings on the same person at intervals of several days gave results that did not vary more than a small fraction of a milligram. The effect of diet was not marked, although the observation that fasting specimens gave slightly higher value was confirmed.[6]

Analogous to this, and contrary to the statement that [7] ingestion of carbohydrate tends to increase the bilirubin content, was the finding in determinations made on patients undergoing glucose tolerance tests: A decrease in bilirubin averaging 0.5 mg. occurred in practically every case, as a rule in the second hour specimen, seldom in the first hour specimen. The method of performing these tests, however, involved giving 1.75 Gm. of glucose per kilogram of body weight in a fairly concentrated solution; and the possibility of the decrease being due to a change in blood volume must be borne in mind.

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6. Broun, G. O.; Ames, O.; Warren, S., and Peabody, F. W.: J. Clin. Investigation **1**:295, 1925.

7. Whipple: Harvey Lecture, 1921-1922, vol. 95.

CLINICAL APPLICATIONS

The quantitative determinations of blood bilirubin, together with the type of reaction given, are of considerable importance in a variety of conditions, from the point of view of diagnosis and prognosis and as an index to the progress of the disease. This is true in cases of damage of various types to the liver, diseases of the gallbladder and biliary tract, diseases of the blood and blood-forming organs, and possibly other conditions in which the significance of the bilirubin is not as yet appreciated. At the same time, a quantitative estimation of urobilinogen in the urine[8] affords a valuable check and confirmatory evidence.

Probably the most important practical application of the determination is, as a routine measure, in patients receiving arsenicals, particularly of the arsphenamine group. Here it affords the earliest possible indication of damage to the liver. If a record of the blood bilirubin value is made at the commencement of the treatment and repeated at regular intervals during the course, one may proceed with safety as long as the reading remains normal. A rise of even a few milligrams is significant, and the patient should be treated accordingly. Any considerable rise calls for the immediate cessation of treatment. Once an arsenical hepatitis is established, determinations of the bilirubin afford a reliable evidence concerning the severity and progress of the lesion. The type of reaction given in these cases is always biphasic, though this is recognized with difficulty if the value is low. The tables illustrate results obtained by this method in various types of cases.

As will be noted from table 1, the bilirubin test in arsenical jaundice gives high results, despite the fact that the disease was marked in only one of the foregoing cases. Incidentally, the highest values that I have seen occurred in two cases of Weil's disease in Toronto General Hospital which are not included in the present series; at the height of the disease the readings were 340 and 370 mg., respectively, estimated according to the method described herein. This is considerably higher than occurs in presumably complete obstruction of the biliary tract.

In the two cases of arsenical dermatitis included in the series, the bilirubin content was not increased.

The patient in case 7 was a woman who two days after delivery showed a slight degree of jaundice without fever and with mild anemia; the jaundice progressed for six or seven days, and then gradually disappeared. In view of the biphasic reaction, a diagnosis of toxic hepatitis was made. The patient in case 8 was a woman with a 4 plus Wassermann reaction, who two years previous to the present admission

8. Wallace, G. B., and Diamond, J. S.: Significance of Urobilinogen in Urine as Test for Liver Function, with Description of Simple Quantitative Method for Its Estimation, Arch. Int. Med. **35**:698 (June) 1925.

had undergone a splenectomy for splenomegaly and severe secondary anemia. She now showed jaundice, enlarged liver and marked anemia. Syphilis may or may not have been the etiologic factor in the condition, which in many respects resembled Banti's disease.

As will be seen, the observations in cirrhosis are variable, depending largely on the type. As a rule, the portal variety shows the direct reaction of obstructive jaundice.

THYROTOXICOSIS

Eight patients with thyrotoxicosis were chosen at random from those under treatment in the wards; all showed typical signs and symptoms of hyperthyroidism with basal rates of from plus 35 to 75. The bilirubin was not over 2 mg. per thousand cubic centimeters in any case,

TABLE 1.—*Results of Tests for Blood Bilirubin in Conditions Involving the Liver*

Case	Diagnosis	Mg. per 1,000 Cc.	Reaction
1	Arsenical jaundice	15.6	Biphasic
2	Arsenical jaundice	83	Biphasic*
		72	Biphasic
		68	Biphasic
3	Arsenical jaundice	36	Biphasic
4	Arsenical jaundice	29.5	Biphasic
5	Arsenical dermatitis	2.5	Indirect
6	Arsenical dermatitis	1.5	Indirect
7	Hepatitis; postpartum	26.5	Biphasic
		18.5	Biphasic
8	Hepatitis; syphilitic	25	Biphasic
		42	Biphasic
		36	Biphasic
9	Hepatitis; syphilis; anemia	4	Indirect
10	Portal cirrhosis	34	Direct
11	Portal cirrhosis	24	Direct
12	Cirrhosis; ascites	1.5	Indirect
13	Cirrhosis; ascites	1	Indirect
14	Cirrhosis; ascites	1.5	Indirect

* Under thiosulphate treatment.

and it was as low as 0.5 mg. The consistently normal values would seem to indicate that the degree of possible damage to the liver, so far as these cases were concerned, is so slight as not to effect the bilirubin values to any discernible extent.

CONDITIONS INVOLVING GALLBLADDER AND BILE DUCTS

The results in cases of cholecystitis and cholelithiasis without obstruction show that the hepatitis that has been stated by various authors frequently to accompany these conditions is not as a rule demonstrable by increase in bilirubin content. In twenty cases of cholecystitis of varying grades of severity—several acute cases but none showing evidence of biliary obstruction—only one gave a reading over 3 mg. (4 mg.). Practically all readings taken individually would be concluded to be normal, and determinations of bilirubin in this condition would appear to have little diagnostic value, particularly in the chronic

stage. It is interesting to note, however, that if the average reading for the group is taken, 1.85 mg., it is approximately 50 per cent higher than that shown by the group of normal persons. That this may be of significance in indicating a slight degree of hepatitis is possible.

Two patients with catarrhal jaundice were seen in this series, giving readings varying from 30 mg. to 15.5 mg. In one case the patient gave a direct reaction at the height of the condition; all other reactions were biphasic.

In cases of common duct stone and occlusion of the common duct by tumors, the bilirubin tests gave varying values, the highest being 142 mg. in a case of carcinoma of the pancreas. The principal value of the determination is in following the degree of the obstruction.

TABLE 2.—*Results of Tests for Blood Bilirubin in Conditions Involving Blood* *

Case	Diagnosis	Mg. per 1,000 Cc.	Reaction
1	Pernicious anemia	19.5	Indirect
2	Pernicious anemia	19.5	Indirect
3	Pernicious anemia	18.5	Indirect
4	Pernicious anemia	16.5	Indirect
5	Pernicious anemia	15	Indirect
6	Pernicious anemia	14	Indirect
7	Pernicious anemia	14	Indirect
8	Pernicious anemia	11	Indirect
9	Pernicious anemia	9	Indirect
10	Pernicious anemia	8	Indirect
11	Pernicious anemia	7	Indirect
12	Pernicious anemia	5	Indirect
13	Pernicious anemia	4.5	Indirect
14	Secondary anemia	3.5	Indirect
15	Secondary anemia	2.5	Indirect
16	Secondary anemia	2	Indirect
17	Secondary anemia	1	Indirect
18	Secondary anemia	1	Indirect
19	Sprue with anemia	3	Indirect
20	Hemolytic jaundice	3.5	Indirect
21	Splenic anemia	5	Indirect
22	Splenic anemia	7	Indirect
23	Hemolytic anemia, pregnancy	14.5	Indirect
24	Purpura hemorrhagica	2	Indirect
25	Paroxysmal hemoglobinuria	2	Indirect

* These represent maximum values obtained in each case.

In table 2 are given the maximum values obtained in the individual cases seen in the wards over a period of some months. Many of these cases gave considerably lower readings on various occasions, indicating the value of repeated determinations. It has been pointed out by many observers [9] that the blood in pernicious anemia has an increased bilirubin content, although not to the extent shown in the foregoing series. This increase can often be surmised from the appearance of the serum alone, deceptive as this frequently is. It will be noted that as a rule the variation in values between primary and secondary cases of anemias is distinctly marked. The borderline case with values of from 3 to 5 mg. per hundred cubic centimeters presents the greatest difficulty. A case of primary anemia undergoing a remission will give values as low or

9. Fishberg, A. M.: Am. J. Med. Sc. **172**:81 (July) 1926.

lower than this. Here repeated estimations over a period of time are necessary, and too much reliance must not be placed on the bilirubin value as a means of diagnosis. Any reading over 3 mg. however, should render one suspicious of a hemolytic factor.

Determinations at regular intervals also constitute a valuable means of determining the progress of a case, effect of treatment and other factors. The occurrence of a remission will be indicated by a decrease in bilirubin some days before any definite change will occur in the blood count other than an increase in the reticulated cells. The course of a typical case (case 4) is shown in table 3.

Case 23 in table 2 represents an interesting case of anemia in pregnancy. On admission, at the stage of seven months, the patient showed

TABLE 3.—*Variations in Bilirubin in Case 4*

Date	Hemoglobin	Red Blood Cells	Mg. per 1,000 Cc.	Remarks
11/20/26	30	1,110,000	15.6	
12/ 3/26	25	1,120,000	16.5	Feeding with spleen commenced
12/ 8/26	25	1,100,000	15.5	
12/14/26	25	.....	15	
12/17/26	..	.....	....	Regular high liver diet instituted
12/21/26	25	980,000	14.5	
12/23/26	30	1,310,000	10.5	
12/28/26	33	1,840,000	7.5	
1/ 4/27	40	1,990,000	5.5	
1/19/27	48	2,490,000	2.5	

TABLE 4.—*Results of Tests for Blood Bilirubin in Lead Poisoning*

Case	Mg. per 1,000 Cc.	Remarks
1	1.5	Mild symptoms; trace of lead in urine
2	2	Mild symptoms; history of exposure; no lead in urine
3	4.5	Moderate symptoms; lead in urine
4	5	Moderate symptoms; lead in urine

a severe anemia, primary in type, and required several transfusions before term. The bilirubin value, which had persistently been about from 12 to 14 mg. per thousand cubic centimeters, fell to 7 mg. within four days of delivery and thence to normal.

Case 25 was one of paroxysmal hemoglobinuria in which an effort was made to demonstrate an effect on the blood bilirubin of exposure to cold by testing blood before and one hour after immersion in cold water. No perceptible change was noted; neither, however, was hemoglobinuria produced.

The occurrence of jaundice of various grades in lead poisoning has long been a subject of speculation. In 1839, Tanquerel reported fifty-one cases of clinical jaundice in 1,217 cases of lead colic. This jaundice has been variously ascribed to direct hemolytic action of lead on the red cells and also to lesions in the bone marrow. Heubel, in 1871, obtained evidence of hemolysis in these cases by finding increased pig-

ment in the serum, bile and urine. The direct action on the red cells has been more recently demonstrated by Aub, Fairhall and others.[10] It is most probable that this is the cause of the constant mild secondary anemia in lead poisoning. The small series in table 4 would appear to indicate the possibility of determinations of bilirubin being of some value in the diagnosis of this condition. Further studies in this connection on a larger series would be of interest.

SUMMARY

A study was made of the blood bilirubin values in the normal person and in a series of cases representing various conditions in which abnormality in this regard might be expected. The technic used was that of Thannhauser and Andersen, with some slight modifications which I believe offer greater convenience and accuracy.

The normal range was from 0.5 to 3.5 mg. per thousand cubic centimeters of blood. The effect of race and sex was not apparent, and that of pigmentation was slight. Fasting specimens gave slightly lower results.

I believe the determination to be of considerable value in patients receiving arsenical preparations as giving the earliest possible indication of damage to the liver. In cases of disease of the liver, important information is often given, but the type of reaction must not be entirely relied on as a means of diagnosis. Characteristically, there is no discernible effect in cases of cholecystitis and cholelithiasis without obstruction, twenty cases giving results which were within normal range, with one exception. The bilirubin is a valuble adjunct to the diagnosis and prognosis in diseases of the blood and blood-forming organs.

That the estimation may be of value in other conditions I believe possible, and in this class I would include lead poisoning, where the results warrant further investigation.

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10. Aub, Fairhall, Minot, Reznikoff: Medical Monographs no. 7.

PERNICIOUS ANEMIA AND TROPICAL SPRUE

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Because of some recent papers suggesting, and in some cases even stating, that tropical sprue and pernicious anemia are the same disease, it has seemed worth while to give some of our observations on cases of sprue, especially comparing those that are typical of the latter condition. The identity of these two diseases has been particularly insisted on by Wood,[1] who has accepted Ashford's theory that *Monilia* causes sprue, and who has been able to take cultures of this yeast in all of his recent cases of pernicious anemia. He quite rightly says that for those who believe *Monilia psilosis* is the cause of sprue, the finding of this organism in pernicious anemia is one more point tending to prove the identity of these two diseases. We have recently published our results [2] with mouth and stool cultures in eleven cases of sprue and seventeen cases of pernicious anemia. Recently also Reed and Wyckoff [3] have written on the similar changes found in sprue, pernicious anemia and combined degeneration of the spinal cord. They have not been able to grow *Monilia* in their cases of sprue; they have found the blood picture of pernicious anemia and evidences of degenerative changes in the spinal cord. In a paper on the achlorhydria family tree of diseases, Christian [4] gives the similar observations in sprue and pernicious anemia; he suggests that these diseases may be the same and also that sprue may develop into pernicious anemia. Bramwell [5] has listed the similarities in the physical observations and blood picture, but admits that there is not the emaciation in pernicious anemia that there is in sprue. Priston [6] has written on the similarity of the appearance of the tongue in the two conditions. Van der Scheer [7] stated that the presence of hydrochloric acid in sprue would differentiate the two diseases.

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1. Wood, E. J.: Pernicious Anemia in Its Relationship to Sprue, Am. J. Med. Sc. **159**:28, 1925.
2. Baumgartner, E. A., and Smith, Glenn, D.: Monilia Psilosis as a Cause of Tropical Sprue, Am. J. Trop. Med. **6**:433 (Nov.) 1926.
3. Reed, Alfred C., and Wyckoff, H. A.: The Common Picture of Sprue, Pernicious Anemia, and Combined Degeneration, Am. J. Trop. Med. **6**:221, 1926.
4. Christian, H. A.: The Achlorhydria Family Tree of Diseases, Northwest Med. **24**:531, 1925.
5. Bramwell, Sir B.: Sprue and Pernicious Anemia, Brit. M. J. **1**:365, 1924.
6. Priston, J. L.: The Tongue in Addison's Anemia, Brit. M. J. **1**:216, 1924.
7. Van der Scheer, A.: Sprue and Pernicious Anemia, Nederl. Tijdschr. v. Geneesk. **2**:1468, 1924; abstr., J. A. M. A. **83**:1548 (Nov. 8) 1924.

Hampson and Shackle,[8] under a title of megalocytic and nonmegalocytic anemias, have found that in these two diseases, as well as in bothriocephalus anemia, the red cells are much larger than in many other diseases. Elders[9] also has written concerning the similarity of the anemia in the two conditions.

DIAGNOSIS

Although pernicious anemia has been studied and written about a great deal, there is probably some difference in opinion as to what is requisite for a diagnosis. Hurst[10] has recently taken up this subject, especially the emphasis to be placed on the presence or absence of hydrochloric acid in the gastric contents in this disease. From a series of cases of pernicious anemia from the literature and from his own experience he finds that in 98.3 per cent of the cases there is achylia, although he concludes that the diagnosis of pernicious anemia is not justified when hydrochloric acid is present. Levine, as well as several other writers, has found hydrochloric acid in several of his cases of pernicious anemia. If such cases can be accepted as pernicious anemia, then at least one if not two of the cases recently discussed as atypical anemia by Munford[11] should be diagnosed as pernicious anemia. So far our clinic has not been ready to accept that diagnosis in cases in which hydrochloric acid was found.

The question of anemia, of course, arises more often than any other, and although the blood picture of pernicious anemia is well known, it may be well to list again the points that are generally accepted. There is a low red cell count, often below 3,000,000, and a relatively high hemoglobin content, giving a color index over 1. Poikilocytosis, a marked variation in the size of red cells and especially the occurrence of large red cells, polychromatophilia, stippling and megaloblasts or large nucleated red cells are found on examining the blood smear; a low white cell count, a high lymphocyte percentage and a low platelet count complete the usual picture. Recently again an article has appeared by Faber and Gram[12] suggesting that they do not consider a high color index so essential to a diagnosis of pernicious anemia as do some others.

Another difficulty in comparing the two diseases is that the cause of pernicious anemia is not yet known, and many have not accepted

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8. Hampson, A. C., and Shackle, J. W.: Megalocytic and Nonmegalocytic Anemia, Guy's Hosp. Rep. **74**:193, 1924.

9. Elders, C.: Tropical Sprue and Pernicious Anemia; Etiology and Treatment, Lancet **1**:75 (Jan. 10) 1925.

10. Hurst, Arthur F.: The Pathogenesis of Subacute Combined Degeneration of the Spinal Cord with Special Reference to Its Connection with Addison's (Pernicious) Anemia, Achlorhydria and Intestinal Infection, Brain. **48**:218, 1925.

11. Munford, S. A.: Atypical Anemias, Clifton M. Bull. **11**:139, 1925.

12. Faber, K., and Gram, H. C.: Relations Between Gastric Achylia and Simple and Pernicious Anemia, Guy's Hosp. Rep. **74**:193, 1924.

Ashford's views that *Monilia psilosis* is the cause of sprue. Smith [13] has recently given some experimental proof of the latter view. Wood's [1] finding of *Monilia* in his cases of pernicious anemia has been noted. In our first report of bacteriologic observations on eleven cases of sprue and seventeen cases of pernicious anemia we showed that this organism was often present. It was also found in five of eighteen other cases with diarrhea.

The pathology of pernicious anemia is better known than is the cause. It has long been known that irregular small ulcerations occur in the mouth in sprue. These appear to be acute ulcers, from which some experimenters have grown *Monilia*. Ulcerations in the lower part of the digestive tract have been described, but less is known about them. The thinning of the wall of the digestive tube has been mentioned again and again, but we have found few autopsy reports to confirm this.

As Wood has stated, the distinguishing characteristics of sprue and pernicious anemia are not always well marked. Ordinarily in cases of pernicious anemia there is a fairly typical history of loss of appetite, sore tongue, no special loss of weight and certain neurologic symptoms. Clinically, a smooth or inflamed tongue, a lemon-tinted skin in a fairly well nourished patient and various neurologic observations of combined degeneration of the spinal cord are found. The changes in the spinal cord have been said to occur in about 80 per cent of the cases (Waltman, 1924). With achylia and a more or less characteristic blood picture, the diagnosis of pernicious anemia is justified.

In a pronounced case of sprue the patient will usually have diarrhea or the history of diarrhea with large frothy stools, ulcers or the history of ulcers of the mucous membrane of the mouth, marked loss of weight, and weakness and neurologic symptoms like those in pernicious anemia and sometimes like those in tetany, which is said to occur later in about one fourth of the cases. These patients are emaciated, have ulcers of the mouth, a distended abdomen and, occasionally, evidence of tetany. There may be an addisonian-like anemia and achylia.

DISCUSSION OF CASES

We have therefore again studied the records of our cases of sprue with the idea of comparing them with the observations in cases of pernicious anemia, more particularly those made in a recent series of fifteen cases the records of which are more complete than those of the earlier cases. Some of these patients have been observed over a period of several months; some of them for two years. The records have been studied closely for the blood pictures and results of gastric examinations, as we possibly rely more on these two tests than on any other for laboratory aid in the diagnosing of pernicious anemia.

13. Smith, I. W.: Monilia Psilosis Ashford in Severe Anemia Associated with Sprue Syndrome, Philippine J. Sc. **24**:147, 1924.

The changes found in the blood in pernicious anemia have been stated in the foregoing. Nine of the fifteen cases of sprue (table) had a color index over 1, but five of these showed an index less than 1 before the patients left our hospital. The highest color index was 1.4, and this patient showed a color index of 0.7 when discharged. Four of the patients had a count below 2,000,000 red cells and nine below 3,000,000. In the table, in the column for erythrocytes (column 2), in which two figures are given, they represent the lowest and highest figures obtained for that patient. As a rule, the lowest figure is the one found on entry, while the highest is the one obtained before the patient left. In the column for color index two figures for each case are shown only in patients in whom there was either a change in the color index or a marked change in the red cell count. The second figure of the two represents that obtained at the latest count. During observation here, seven of the patients had nucleated red cells, but megaloblasts were found only in the patient that died. Changes in the red cells were found in eleven cases and were marked in four (indicated by ++ in the table). Anisocytosis appears to be less marked but actual measurement of the cells in two cases (cases 4 and 6) showed the curve (Hampson and Shackle) of the cell diameters to be to the right of that found for normal cells. Eight of the cases showed a low number of leukocytes at some time, but in only four cases was the number of leukocytes low in the majority of the counts, while in three of these four cases and in five others there were over 40 per cent of lymphocytes in a majority of the counts. In nine cases the platelets were counted; in six, they were less than 100,000, which is definitely lower than normal.

Three of the fifteen patients with sprue did not show free hydrochloric acid in gastric fractional tests (cases 3, 8 and 9). On a fourth patient we did not make a test, but free acid was not present in the contents of the stomach at postmortem examination (case 1). Four months previously, an Ewald test at another clinic had showed a free acidity of 19 and a total acidity of 28 per cent in this case. In a fifth case (case 6) the patient refused any gastric examination; so our only method of testing for acidity was by means of the alkaline tide, which was negative. Hubbard [14] has shown that this test confirms either the presence or the absence of acidity in about 80 per cent of his cases, and we, therefore, accept this patient as being achylic. In this series of cases of sprue, then, there are five (33 per cent) with achylia which could be diagnosed pernicious anemia as far as this observation is concerned.

Other laboratory tests which may indicate pernicious anemia are tests for bile pigments, urobilin and urobilinogen of the urine or feces.

14. Hubbard, R. S., and Munford S. A.: Alkaline Tide in Achlorhydria, Arch. Int. Med. **35**:576 (May) 1925.

Such tests were not performed on the feces, but in eight cases twenty-four hour specimens of urine were examined. Only three of these showed a definite increase in these compounds.

A recently described laboratory observation in some cases of sprue is a low blood calcium. This, as several investigators have shown, is often associated with a clinically demonstrable tetany. In four of the cases of sprue the calcium content was 7.3 mg. per hundred cubic centimeters of blood or less, which is well below normal; in four others the blood calcium was from 8.3 to 8.6, which most investigators will agree is below the accepted normal. The blood fat was low in three cases tested and the blood cholesterol in two.

In the fifteen cases of sprue studied there has been a history of ulcers of the mouth. Besides this, two patients have complained frequently of a dry mouth. This caused a great deal of discomfort to one patient, and there was a history of ulcers early in the course of the disease which was all but forgotten. Six of the patients had ulcers in the mouth or on the lip during observation here, but three complained of a burning tongue when nothing unusual could be seen. In nine cases the tongue was definitely normal, while in cases 1, 4, 8 and 9, it somewhat resembled the tongue in pernicious anemia. The table also shows that there was loss of weight in all cases, and in seven this amounted to between one half and one fourth of the average weight.

Definite reactions showing tetany have been observed in only three cases. One of these (case 4) patients showed the tetany spasm on entry; in this case and in two others (cases 6 and 8) there were positive Trousseau and Chvostek signs. Of four patients tested with the electric battery, only one (case 4) gave the positive reactions of tetany, although one other gave suggestive reactions.

The reports on stool and mouth cultures in eleven of the cases of sprue and other conditions have been published elsewhere. In the last column in the table it can be seen that stool cultures were positive in all cases of sprue except in case 14. In case 6 the first culture produced atypical *Monilia* which did not produce gas in maltose. Several cultures were made from the stools of case 13 before *Monilia* was obtained.

There have been listed also fifteen cases of pernicious anemia in which the various observations were compared with those in sprue. In only eight of our cases of pernicious anemia have tests been performed to determine the blood calcium. In one case the blood calcium was 8.6 mg. per hundred cubic centimeters of blood, which was the only case in which this constituent was below normal. The remainder ranged in the normal limits from 9 to 11 mg. per hundred cubic centimeters of blood. Stool culture showed that in five cases *Monilia* was grown, one of which was atypical in its reactions on the sugars. In one case *Monilia* was grown from the gastric contents obtained for fractional analysis.

A. Cases of Sprue

Case	Red Blood Cells (in Millions)	Color Index	Red Cell Changes	Platelets	Gastric Acidity	Possible Pernicious Anemia	Tetany			Loss in Weight	Neurologic Observations	Blood Calcium	Monilia Culture
							Electric	Chvostek	Trousseau				
1	2—	1+	++	23,000	None (autopsy)	Yes	.....	.....	.....	Severe 155 to 80	Negative	5.6	Tongue + Stools +
2	4±	1—	None	.....	Low normal	....	.....	Negative	Negative	115 to 98	Negative	8.6	Stools +
3	2— 3.5+	1+ 1—	++ 0	98,000	None	Yes	Negative	.....	.....	Severe 150 to 98	Negative	8.3	Stools +
4	2 3.5+	1+ 1—	+ 0	68,000	Normal	....	Positive	Positive	Positive	Severe 110 to 67	Negative	6.2	Stools +
5	4±	1—	±	152,000	Normal	....	.....	.....	.....	170 to 133	Negative	8.4	Stools +
6	2±	1+	+	34,000	Negative tide	Yes	.....	Positive	Positive	160 to 104	Negative	7.3	Stools +
7	3— 4+	1+ 1—	±	285,000	Normal	....	.....	.....	.....	124 to 90	Negative	11	Stools +
8	2— 3+	1+ 1—	++ +	107,000 74,000	None	Yes	.....	Positive	Positive	130 to 108	Negative	3.1	Stools +
9	2 3±	1+ 1+	++	.....	None	Yes	.....	Negative	Negative	160 to 127	Not done	10.3	Stools +
10	4+	1—	None	239,000	Normal	....	.....	Negative	Negative	170 to 147	Negative	8.6	Stools +
11	3±	1+ 1—	+	60,000	Normal	....	.....	Negative	Negative	180 to 132	Not done	10.4	Stools +
12	4+	1—	None	.....	Normal	....	Negative	Negative	Negative	125 to 99	Negative	9.0	Stools ±
13	2 4.6	1+ 1+	+	.....	High normal	....	.....	.....	.....	150 to 90	Negative	9.6	Stools +
14	4+	1—	None	.....	Normal	....	.....	.....	.....	200 to 160	Negative	11.3	Stools —
15	4.5	1—	None	.....	High normal	....	.....	.....	.....	160 to 147	Not done	...	Stools +

B. Cases of Pernicious Anemia

1	2±	1+	++	.....	Negative tide	Yes	.....	.....	.....	.....	Positive	...	Stools +
2	2/3	1+	++	.....	None	Yes	.....	.....	.....	130 to 104	Positive	8.7	Stools —
3	2±	1+	++	.....	None	Yes	.....	.....	.....	150 to 114	Positive	10.2	Stools + Mouth —
4	2+/4	1+	++	.....	None	Yes or subacute combined degeneration	.....	.....	.....	140 to 120	Marked	...	Tongue —
5	3/4	1+	+	.....	None	Proved autopsy	.....	.....	.....	127 to 117	Positive	...	Stools — Mouth —
6	3—	1+	+	.....	None	Possible cancer (autopsy elsewhere)	.....	.....	.....	75 pounds loss	Positive	...	Stools +
7	3±	1+	+	.....	None	Yes	.....	.....	.....	.....	Positive	...	Mouth —
8	1+	1+	++	.....	Ewald none	Yes	.....	.....	.....	150 to 135	Positive	11.9	Stools +
9	2—/3	1—/1+	++	.....	None	Yes	.....	.....	.....	147 to 135	Negative	10.7	
10	2+	1+	++	.....	None	Yes	.....	.....	.....	130 to 120	Very slight	...	Stools
11	3—	1—	+	.....	None	Yes	.....	.....	.....	150 to 145	Positive	...	Gastric contents +
12	1+/4	1+/1—	++/+	.....	None	Yes	.....	.....	.....	.....	Slight	...	Stools — Mouth —
13	3—	1+	++	.....	None	Yes	.....	.....	.....	None	Positive	9.2	Stools —
14	2+/3	1+	++	.....	None	Yes or subacute combined degeneration	.....	.....	.....	.....	Marked	10.3	Stools —
15	3+/3+	1—/1+	+	.....	None	Yes	.....	.....	.....	.....	Marked	9.0	Stools + Mouth +

COMMENT

Several articles have appeared within the last few years in which various authors have tried to show that pernicious anemia and sprue are one and the same disease. Wood, in this country, has probably been the most insistent on this question. In sixteen cases of pernicious anemia he has been able to grow *Monilia psilosis* on cultures taken from either the mouth or the stools. We have taken cultures from the tongue or the mouth in seven cases of pernicious anemia, and so far have been successful in growing this organism in one (case 15). In our cases of sprue we took cultures from the sore tongue (case 1) or from definite ulcers (five cases); here also we failed, except in the first case, in which cultures were taken twice and were positive both times. Cultures from the stool in the cases of sprue were positive at the first attempt in thirteen of our cases, although one (case 6) did not yield an organism giving the accepted typical sugar reactions at the first culture. In one case (case 14) in which the patient had not had definite symptoms for eight years, only one culture was made, and typical *Monilia psilosis* was not found. In eight attempts in another case (case 12), *Monilia* was obtained only twice, one of which was atypical in maltose. In case 13 five cultures have been negative and only once was typical *Monilia* obtained. Our attempts at growing *Monilia* from the cases of pernicious anemia have been less successful, although this was not undertaken until we had had considerable experience. As is shown in another article, an organism giving the typical cultural reactions of *Monilia psilosis* was found in cultures from the feces in only three of seventeen cases, while in one, this organism was grown from the gastric contents. In the table we have added one more case (case 15) in which a positive culture was obtained.

If one would accept *Monilia* as the causative agent of sprue, and, following Wood, also of pernicious anemia, then our results would at least indicate that the organism in the latter condition is less easily grown. Our technic and the mediums used for the two conditions were the same.

Many have called attention to the similarity of the anemia in the two diseases. Our figures show that as far as the color index is concerned, and this is the most frequently stressed point in the blood picture, it was over 1 in nine of our cases of sprue, but in five of these it became 1 — during the patient's stay in the hospital. So far, only one of our cases of pernicious anemia has shown this (case 11). Van der Scheer [7] also found a high color index. Elders has given figures of blood counts in cases of sprue calling attention to a 1+ index. However, his quoted figures of erythrocytes and hemoglobin do not bear out his figures of color index, according to our method of calculating. Wood apparently has often found similar blood pictures in the two diseases.

In only one of our cases of sprue have we found megaloblasts. If one would lay stress on the blood picture, megaloblasts and large red cells are the most characteristic of pernicious anemia. The various changes in the red cells enumerated in the foregoing have certainly not been as marked in our cases of sprue as we would demand for criteria in diagnosing pernicious anemia. Various British investigators, especially Hampson and Shackle, have, following Jones'[15] description, measured the red cells in various diseases, and have plotted curves of the number and the diameter of these cells. They found that in sprue, pernicious anemia and bothriocephalus anemia the curves show that the red blood cells are larger than those in normal cases and in many other diseases. The red cells were measured in two cases of our series (cases 4 and 6). Both showed many large cells, the largest measuring well over 8 microns, thus confirming the resemblance between the red cells of sprue and those of pernicious anemia. Bastedo and Famulener[16] found a high color index in two of their cases of sprue in which the patients died. In his cases of sprue Smith has described the typical blood count of pernicious anemia, but he has failed to find the cell changes characteristic of pernicious anemia. He mentions the lack of study of the bone marrow in sprue. In the case described by Baumgartner and Thomas[17] (case 1), the bone marrow was certainly not typical of pernicious anemia, although the blood count and the postmortem achylia might suggest that diagnosis.

That there may be a profound anemia in sprue, no one doubts. Several of our patients have been severely anemic. That this helps to confuse the picture, we also admit. Wood's statement is also true, that the blood picture may be misleading, so that one disease may be mistaken for the other. Depending too largely on the presence of anemia and achylia, we, too, have wrongly diagnosed at least one case as pernicious anemia; one must not depend too much on the blood picture in diagnosing this condition. We believe that the anemia in sprue is, as Smith[13] found, aplastic. A high color index is not unusual in our series, but in most cases this became 1 — as the patient improved, a change also described by Elders. Recently Gram and Faber[12] have found this occasionally in pernicious anemia, but in our experience it is infrequent in the latter group.

If we accept achylia as a necessary observation in the diagnosis of pernicious anemia, then only five of our cases of sprue could be considered pernicious anemia (table). These five achylic cases had blood pictures that might readily be mistaken for the picture in pernicious

15. Price-Jones, C.: Anisocytosis Special Reference to Pernicious Anemia, Guy's Hosp. Rep. **74**:10, 1924.

16. Bastedo, W. A., and Famulener, L. W.: Tropical Sprue, J. A. M. A. **81**:2102 (Dec. 22) 1923.

17. Baumgartner, E. A., and Thomas, W. S.: A Case of Tropical Sprue with Autopsy, Clifton M. Bull. **11**:90, 1925.

anemia. As a matter of fact, in three of our other cases of sprue there was almost as severe an anemia, but in these the presence of free hydrochloric acid would rule out pernicious anemia. Priston [6] stated that hyperacidity or normal acidity is found in sprue; unfortunately, this is not always true, at least it has not been true in our cases unless we have wrongly diagnosed the five achylic cases. Bastedo and Famulener found that in most of their cases of sprue acidity was normal, although two patients that died were achlorhydric.

Achylia is almost always found in pernicious anemia. Faber and Gram believe that there was free hydrochloric acid in the stomach in five of their fifty-two patients with pernicious anemia. Levine reported three such cases. As stated, Hurst found that in 98.3 per cent of the cases of pernicious anemia which he collected from the literature the patients had achylia.

If the entire blood picture and the achylia are considered, it must be admitted that five of our cases might be called pernicious anemia. We admit with Wood that any one without experience with sprue could readily fall into this error; even with some experience, we have done so in this clinic. Such an error occurs when only these two observations are used in diagnosing these conditions.

Although eight of our cases of sprue have had a low blood calcium, only four of these have been decidedly low, while in four it was no lower than in one of our cases of pernicious anemia. As we did not know of any published figures giving the blood calcium in cases of pernicious anemia, we studied a few cases, and we found that this varied from 9 to 11.9, except in one case with a blood calcium of 8.7, which a week later was 10.4. Three of the five cases of sprue mentioned in the foregoing as possible cases of pernicious anemia had a low blood calcium, which, according to our figures, would be against the diagnosis of pernicious anemia.

The loss of weight in sprue is often great. In half of our cases the loss was from one fourth to one half of the patients' average weight. The greatest loss noted was in three of the five cases that from the achylia and blood pictures could be called pernicious anemia. We believe with Bramwell that usually there is not the marked emaciation in patients with pernicious anemia that one finds in patients with sprue, and the results in our table certainly do not show that there was a marked loss. Probably all will agree that there is usually little loss of weight in patients with pernicious anemia; and yet this cannot be taken as an infallible observation, as we have recently found. A markedly emaciated patient entered the clinic with a diagnosis of sprue. A blood count and achlorhydria indicating pernicious anemia were found. No neurologic symptoms and no changes in the spinal cord were found on physical examination, nor were there any signs of tetany. What

was the diagnosis to be? The patient came from a region in which sprue is found, which further complicated the picture. The severe loss in weight we believed to be more characteristic of sprue. There was not the typical smooth tongue of pernicious anemia. The blood calcium was normal. Some blood serum was then tested by Macht, who has described a method demonstrating a toxic substance in the blood serum in cases of pernicious anemia.[18] This test showed a marked toxic substance in this patient.

Neurologic examinations have been made in almost all of our cases of sprue. Eight patients had the symptoms of numbness and tingling complained of in pernicious anemia. In one case (case 1) definite physical observations of degeneration of the posterior column was said to have been found at another clinic. In none of these cases have we found any definite changes indicating degeneration of the posterior column, and in the one case that came to autopsy (case 1) no changes in the spinal cord were found with the special stains used. But other nerve observations are sometimes present in sprue. Tetany was first described in this country in one of Bovaird's[19] cases by Barach and Murray.[20] We found evidences of tetany in three of our cases. So far as we know, this has not been described and is not present in pernicious anemia; yet two of these cases of tetany are among the five that may possibly be pernicious anemia, and in these two and in one other of the five there was a low blood calcium.

Mental depression is usually pronounced in sprue, and this has been rather infrequent in our patients with pernicious anemia. One of our cases of sprue (1913) was diagnosed neurasthenia for some months before the true condition was suspected. The numbness and tingling in the two diseases are almost similar, so that one wonders why degeneration of the posterior column, which occurs in pernicious anemia, is not found in sprue. Possibly when more necropsy material is available, such changes will be found. Parathyroid changes as evidenced by tetany were not found in case 1, although the patient had a blood calcium of 5.6 mg. which at postmortem examination was found to be 5.3 per hundred cubic centimeters of blood.

Clinically patients with sprue have a different history from that of patients with pernicious anemia. The diarrhea in our cases of sprue was much more marked and constant than in our cases of pernicious anemia. Loss of strength and mental depression are more severe than we have noted in pernicious anemia. The type of diarrhea, loss of

18. Macht, D. I.: A Study of the Toxin of Pernicious Anemia, Proc. Soc. Exper. Biol. & Med. **23**:209, 1925.

19. Bovaird, D.: A Study of Tropical Sprue or Psilosis, J.A.M.A. **77**:753 (Sept. 3) 1921.

20. Murray, A. L., and Barach, A. L.: Tetany in a Case of Sprue, J. A. M. A. **74**:786 (March 20) 1920.

weight and strength without a blood picture of pernicious anemia in one case made a differential diagnosis from pancreatic carcinoma difficult. The diarrhea in sprue often occurs in the early morning. The large, gray, frothy stools are characteristic, but may occur in pancreatic disease. One patient, case 3, returned in 1925 markedly underweight and severely anemic, but with fairly normal stools and without diarrhea; in case 7 the stools were not typical for a long time.

Clinical observations in the two conditions must be taken into consideration with the laboratory tests. As Wood has stated, the latter is too often stressed. We believe the severe loss in weight is characteristic of sprue, and we did not find this in our cases of pernicious anemia, except in the one referred to in the foregoing. A constant diarrhea, and especially the early morning diarrhea, is not often seen in pernicious anemia. The characteristic appearance of the tongue in cases of pernicious anemia has not been present in our cases of sprue. The small liver stressed by some as present in sprue was not especially notable in our series.

Differentiation of the two diseases is sometimes difficult. The unknown etiology of one and the not generally accepted theory of *Monilia psilosis* as a cause of the other gives little aid in distinguishing them. Wood's finding of *Monilia* in pernicious anemia confuses the picture still more. In our experience the almost constant finding of *Monilia* in sprue and inconstantly (20 per cent) in pernicious anemia and other conditions (25 per cent) lends some support to Ashford's views that *Monilia* is the cause of sprue, and fails to confirm Wood's contention that these two diseases are caused by the same organism. The presence of a toxic substance in the blood in pernicious anemia patients, as Macht has recently shown, may help to distinguish the two conditions or prove them identical diseases, and may also help in differentiation from the other confusing disease, subacute combined degeneration.

SUMMARY

We have discussed various observations, both laboratory and clinical, in patients with sprue or pernicious anemia. In fifteen cases of the former we found five with achlorhydria and with an anemia similar to that of pernicious anemia—high color index—but possibly with less marked changes in the red cells, certainly fewer nucleated red cells. Three of the cases have had clinical evidences of tetany and a low blood calcium, an observation not yet described in pernicious anemia. None of the patients have had any evidence of degeneration of the posterior column while under our care, although several have had the marked numbness and tingling so frequent in pernicious anemia; while in fourteen of the fifteen *Monilia* has been grown from the stools, and this has occurred only four times in seventeen cases of pernicious anemia.

In a severely emaciated patient who had had marked numbness and tingling similar to that in pernicious anemia, as well as a similar blood picture, the spinal cord did not show combined degeneration and the bone marrow was definitely aplastic; yet the liver and kidney showed the iron pigment characteristic of pernicious anemia.

We do not feel that in our series the two conditions should be diagnosed as the same disease. We do not believe, as Christian suggests, that sprue is late pernicious anemia or the reverse, nor that even in late cases of sprue achylia necessarily develops.

In less than half of the cases of sprue in our series achlorhydria occurred. This is present in the more severe cases in which the anemia is more like that of Addison's type. Many of these patients lose a relatively large amount of weight, from one-fourth to one-half their average weight; some, probably all, experience severe numbness and tingling, and some have definite clinical tetany with a low blood calcium. Some cases probably cannot be correctly diagnosed, at least by methods now available.

HYPOGLYCEMIA AND THE TOXIC EFFECTS OF INSULIN *

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Ever since the appearance of the first publication describing the group of symptoms accompanying overdosage with insulin, these symptoms have been generally considered to be associated with an abnormally low concentration of the blood sugar. A number of papers have subsequently appeared which report instances of spontaneous hypoglycemia accompanied by symptoms similar to those of insulin poisoning. These reports have served to strengthen the assumption that the hypoglycemia and the symptoms of insulin poisoning stand to each other in the relation of cause and effect. Hypoglycemia associated with pallor, muscular twitching, moist skin, irritability, collapse and stupor has been observed by Levine, Gordon and Derick [1] in Marathon runners following severe exertion. Harris [2] has reported hypoglycemia in two nondiabetic patients, associated with hunger, weakness and nervousness, which came on an hour before meals. The blood sugar value in these cases was from 60 to 70 mg. per hundred cubic centimeters. If food was taken at intervals of not longer than from three to five hours, no such symptoms were observed. Hartman and Reiman (mentioned by Jonas [3]) described a patient who experienced severe hunger three or four hours after meals, associated with unconsciousness. A subnormal blood sugar concentration was found, and the symptoms were relieved by food. Liu and Chang [4] have reported one of my cases, that of a man who experienced symptoms typical of mild overdosage with insulin during an attack of severe diarrhea associated with hypoglycemia. Holman [5] has reported similar phenomena following thyroid operations, although the clinical symptoms as described and their association with severe thyroid intoxication make the interpretation rather uncertain. John [6]

* From the Medical Clinic of the Johns Hopkins Hospital and University.

1. Levine, S. A.; Gordon, B., and Derick, C. L.: Some Changes in the Chemical Constituents of the Blood Following a Marathon Race, J. A. M. A. **82**:1778 (May) 1924.

2. Harris, Seale: Hyperinsulinism and Dysinsulinism, J. A. M. A. **83**:729 (Sept. 6) 1924.

3. Jonas, L.: Hypoglycemia, M. Clin. N. Amer. **8**:949 (Nov.) 1924.

4. Liu, S. H., and Chang, H. C.: Hypoglycemia: Report of a Case Unassociated with Insulin Administration, Arch. Int. Med. **36**:146 (July) 1925.

5. Holman, E. F.: Hypoglycemia in Exophthalmic Goiter, Bull. Johns Hopkins Hosp. **34**:69 (Feb.) 1923.

6. John, H. J.: The Lack of Uniformity in the Insulin Reaction, Am. J. M. Sc. **172**:96, 1926.

states that in eight normal persons during tests for glucose tolerance values between 31 and 57 mg. per hundred cubic centimeters were found, always associated with extreme hunger. Josephs [7] has reported toxic phenomena after fasting and during recurrent attacks of vomiting in children associated with low blood sugar values. In general, children appear to show hypoglycemia on fasting more frequently than do adults. Stenström [8] has recently reported a case of spontaneous hypoglycemia in a lactating woman with tuberculosis and heart disease when fed on a diet of fat and vegetables. Symptoms of weakness, trembling and profuse sweating were observed, with hunger and a drawing feeling in the upper part of the abdomen, which were relieved when a normal diet was again given. The fasting blood sugar fell to 50, 40 and 50 mg. per hundred cubic centimeters, respectively, on the three days during which symptoms were observed. As stated, this case bears a striking analogy to the observations of Widmark and Carlens [9] on the puerperal paralysis of cows, which appears to be a hypoglycemic coma completely relieved by intravenous injection of dextrose.

An interesting association is with scleroderma, an example of which has been reported from this clinic by Dr. Longcope.[10] A low fasting blood sugar content has been observed in other cases of scleroderma, together with hypotension, but neither is invariably present.

The association of severe diabetes with extreme emaciation, inanition, diarrhea, hypoglycemia and coma without administration of insulin has been observed in this clinic, and similar cases have been reported from other clinics.

CASE 1.—A man, aged 46, who had had diabetes for six years, with extreme loss of weight and emaciation, and more recently diarrhea and edema of the feet and ankles, had a blood sugar concentration of 54 mg. per hundred cubic centimeters on admission to the hospital. There were no "hypoglycemic" symptoms. On unrestricted ward diet the blood sugar rose to 286 mg. per hundred cubic centimeters, and sugar appeared in the urine. The diet was modified, and the fasting blood sugar fell to from 54 to 87 mg. per hundred cubic centimeters. The basal metabolic rate was —20. His condition seemed satisfactory until one morning five weeks after admission, when without warning he suddenly lapsed into a semistupor, and within half an hour was in deep coma, with slow respirations and somewhat increased pulse rate. The blood pressure was low: 90 mm. systolic and 70 mm. diastolic. An examination of the blood sugar before death five hours later showed only 17 mg. per hundred cubic centimeters. The intravenous administration of dextrose in this patient failed to elicit any response or the slightest improvement. He was under the care of Dr. N. B. Herman, who will report the details more fully later.

7. Josephs, H.: Fasting as a Cause of Convulsions, Am. J. Dis. Child. **31**:169 (Feb.) 1926; Recurrent Vomiting, ibid. **31**:657 (May) 1926.

8. Stenström, T.: "Spontane" hypoglykämische Reaktion bei stillender Frau, Deutsches Arch. f. klin. Med. **153**:181, 1926.

9. Widmark, E. M. P., and Carlens, O.: Ueber die Blutkoncentration bei Kühen, Biochem. Ztschr. **156**:454, 1925.

10. Longcope, W. T.: To be published.

It is possible that a part of the weakness and asthenia of Addison's disease, which, as is known, is usually associated with hypoglycemia and lowered blood pressure, may be due to a similar cause. Reports on studies of the effect of glucose in the treatment in this condition have not appeared. An interesting case report of an insulin-like reaction possibly produced by suprarenal insufficiency has recently been published by Stenström.[11] Doubtless other conditions with coincident hypoglycemia unassociated with the administration of insulin will be reported in the future. One may conclude that a state of hypoglycemia may occur in a variety of conditions, accompanied by some or all of the following phenomena: Weakness, dizziness, pallor, sweating and hunger, mild disorientation, occasionally syncope and stupor and usually hypotension. Such hypoglycemia cannot be distinguished from that following overdosage with insulin.

On the other hand, hypoglycemia unassociated with any symptoms has been reported repeatedly. Jansen from Müller's clinic[12] in Münich has shown the frequency with which hypoglycemia occurs in the edema of undernutrition. He found a blood sugar concentration between 34 and 70 mg. per hundred cubic centimeters in twelve of twenty-four cases; no acute symptoms occurred in any of these. The syndrome of edema, bradycardia, hypotension, polyuria, severe hunger and atrophy of the thyroid, lasting over a considerable period, bears no clearcut resemblance to reactions from insulin. Hypoglycemia has been reported several times in undernutrition, especially in children, but without symptoms. Gray has reported the results of examinations for blood sugar in 461 normal persons. Sixty-four of these had a fasting blood sugar concentration of less than 70 mg. per hundred cubic centimeters, but they did not have any symptoms. It must be admitted that hypoglycemia of this grade has not often been seen in routine examinations of the blood sugar of fasting patients in this clinic.

For the proper adjustment of insulin dosage in cases of severe diabetes during the past two years, my co-workers and I have made blood sugar determinations on our patients at three or four hour intervals for a twenty-four hour period, in order to ascertain the proper balance between food distribution and insulin. Such determinations are readily made on capillary blood from the ear or finger either by the Hagadorn-Jansen method or by a microtechnic based on the Benedict-Folin-Wu method. We have been struck by the large number of low blood sugar readings, between 75 and 45 mg. per hundred cubic centimeters, in patients with severe diabetes, both in those who have taken

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11. Stenström, T.: Einige Bemerkungen über spontanes hypoglykämisches Coma, Deutsches Arch. f. klin. Med. **152**:173, 1926.

12. Jansen, W. H.: Die Ödemkrankheit, Deutsches, Arch. f. klin. Med. **131**:144, 1920.

insulin for a considerable period and in those who had received it for only a short time. Most of these patients have not had any symptoms of overdosage with insulin.

CASE 2.—A man, aged 27, who had had diabetes for two years, and who had taken insulin for twenty months, was admitted to the Johns Hopkins Hospital with marked acidosis following a severe infection of the throat. He was finally rendered sugar-free on a rather restricted diet and the administration of 15 units of insulin three times a day before meals. An all day blood sugar curve was made, which showed a morning fasting blood sugar of 135 mg., a concentration of 130 mg. at 11:30 a. m. just before lunch, 55 mg. at 5 p. m. and 110 mg. at 9 p. m. Although there were no symptoms of a reaction in the late afternoon, it was deemed best to reduce the noon dose of insulin to 10 units. This was done, and two days later at 4:30 p. m., he had definite symptoms of an insulin reaction, with weakness, pallor and sweating. The blood sugar concentration before the symptoms were relieved by orange juice was 60 mg. per hundred cubic centimeters. It is impossible in such an instance as this to see any direct quantitative relation between the occurrence of the insulin reaction and the blood sugar concentration.

REACTIONS TO INSULIN IN PATIENTS WITH DIABETES MELLITUS

The present report is based on a study of patients with diabetes mellitus who have suffered insulin reactions while under treatment in this hospital during the past four years. Nearly all of the patients whose cases have been recorded in the past two years have been under my personal observation. It should be stated that most of the diabetic patients treated have had cases of great severity, and have been admitted either for regulation of insulin dosage, or because of difficulties in management in the dispensary or because of other serious complications. For this reason large doses of insulin have been required in many cases, and a much larger proportion of serious reactions has been observed than probably occur on the average among the whole group of patients who use insulin.

Toxic reactions to insulin may be conveniently divided into two groups. The first are the mild reactions, usually unassociated with conspicuous nervous or psychic manifestations. The second group comprises the more severe reactions, in which nervous or psychic symptoms largely or entirely dominate the picture. The frequent occurrence of this type of reactions has only recently received attention.

Mild Reactions.—In the milder type of insulin reactions a group of symptoms occurs which is usually characterized by gradual development, and in which the subjective sensations described by the patients are often more pronounced than are the objective observations. The symptoms in this group in the order of their frequency are as follows: a sense of weakness, usually generalized, less frequently referred to the abdomen, ordinarily appears first and is nearly always associated with a feeling of trembling or of actual tremor of the extremities. Combined with this may be a diminution of deep muscle

sense and sense of position, particularly in the legs. A sensation of coldness in the extremities, of numbness of the lips, hands or feet, of hunger, or less commonly of thirst are apt to appear. Tinnitus and diplopia occur in certain patients, the latter often in children. Actual sensations of acute pain have been absent, and the absence of actual premonitory aura such as occur in epilepsy is also noteworthy. The main objective physical observations are pallor and generalized sweating and occasionally a moderate rise in the systolic blood pressure, with a smaller diastolic increase. In mild reactions usually no change occurs either in the rate of the heart beat or of the respirations. Speech may be somewhat slurred and the mental response sluggish, but in most mild reactions there is really little change. The pupils tend to be dilated. It is usually assumed that if proper steps are not taken to end the attack promptly, unconsciousness, collapse or convulsions, may occur. This assumption has been given wide currency, probably by analogy to the results of experiments on animals. No deaths occurred in the present series as a direct result of overdosage with insulin. All of the phenomena described are not usually present in any one person. Of interest, however, is the tendency of the symptoms and the sequence of their development in a given patient to be repeated in later attacks. Flushing of the cheeks is much less common than pallor, but we have repeatedly seen mild reactions, characterized by conspicuous flushing of the face and neck.

Severe Reactions.—Of more serious practical importance is the group of severe insulin reactions associated with nervous or psychic manifestations. Cases of this kind are less widely recognized, although good descriptions have recently appeared in the literature (Leyton,[13] Severinghaus,[14] Labbé [15]). At least two types are recognizable. The commonest type of such reactions among our cases is that associated with behavior more or less closely simulating alcoholic intoxication. Often there is a brief preliminary period in which the patient may appear dazed and tremulous, or in which he may at first have a sense of weakness and dizziness. Sweating is rather less common than in milder attacks. The patient may then become noisy, laughing or crying; there is incoordination of speech, the vision is obscured, and there is a characteristic loss of the inhibitions. The arms may be tossed about wildly, and there is a typical drunken, staggering gait, with resistance to attempts at restraint. Often delusions and ideas of persecution appear, and the resemblance to the excitement stage of acute

13. Leyton, O.: Hypoglycaemia, Proc. Roy. Soc. Med. **19**:37, 1926.

14. Severinghaus, E. L.: Hypoglycemic Coma Due to Insulin Overdosage, Am. J. M. Sc. **172**:573, 1926.

15. Labbé, M.: Les incidents des traitements prolongés per l'insuline, Bull. et mém. Soc. méd. hôp de Paris **23**:1123 (June 25) 1926.

alcoholism may be close. Obsessions and hallucinations may occur abruptly, or indeed without premonitory symptoms.

CASE 3.—A man, aged 26, who had been suffering from severe diabetes for the last eighteen months, was admitted to the hospital in diabetic coma, from which he was restored by the usual measures. One evening a month later, when he was receiving 40 units of insulin a day, he suddenly became noisy and excited about an hour after supper. His face became pale; he tossed about the bed, talking in a loud voice and in a confused, irrational manner. He failed to recognize his wife and sister who were visiting him at the time. The blood pressure was unchanged; the blood sugar concentration was 65 mg. per hundred cubic centimeters. The patient was quickly revived with orange juice, and later he had no recollection of the incident. This train of symptoms was repeated in an almost identical manner on two other occasions.

A characteristic feature of these attacks is the complete amnesia which the patient exhibits after recovery. His recollection is usually retained up to the moment when symptoms appear, and then it is lost completely.

CASE 4.—A man, aged 57, with diabetes of five years' duration had been treated with insulin for the past eighteen months. One night he ate a light supper, having previously taken 12 units of insulin. Half an hour later he complained of cramplike pains in the legs and then vomited. He felt tired, went to bed and promptly fell asleep. At midnight he awoke and began talking loudly, tossing about in bed entirely disoriented, and resisting the attempts of his family to pacify him. His speech was thick and nearly unintelligible. He accused the medical attendant who was called to see him of wishing to do him bodily injury, and resisted all attempts at examination. At this time he was taking 12 units of insulin twice a day. He came out of this confusional state after he received orange juice and sugar, but he was not completely rational for an hour or more. He had no recollection later of what had happened.

Loss of Consciousness.—A more serious type of disorder is that characterized by partial or complete loss of consciousness for longer or shorter periods. This often occurs without warning, or if any warning is given, it may be so brief that the patient does not have time to summon aid. It may occur during sleep, being recognized only in the morning when it is found that the patient cannot be aroused. The attack may be one of semistupor in which patients remain for varying periods, the longest which we have observed being sixty hours. When in this condition, the patient may lie totally unresponsive to any stimuli except pain. Resistance is usually offered, however, to manipulations, such as attempts at venipuncture or the insertion of an infusion needle. The patient is usually pale, and he often lies with the eyes half open, staring vacantly. When the eyes will follow light, coarse nystagmoid movements may be elicited. There is nothing characteristic of the behavior of the deep reflexes, which are usually unaltered. The temperature is frequently subnormal, the pulse and respirations are slowed, and the blood pressure is usually low. Irregularities of respiration have been seen in two of our patients, in one of

whom periodic breathing of the Cheyne-Stokes type was observed for a period of thirty minutes. While the patient is in coma, waxy flexibility of the extremities may be observed, and the whole picture much resembles that of catatonic stupor. When the reaction is prolonged, there may be periods of semiconsciousness with subsequent relapse into the state of stupor. The mental state during these periods of partial consciousness is somewhat difficult to define, but it is characteristic. The patient is dull and appears dazed, but may at times recognize persons and places. These occasional glimpses of rationality may deceive the observer into believing the patient's behavior a sham or into minimizing its seriousness.

From such severe psychic reactions or even actual unconsciousness, patients may recover and become rational suddenly, or there may be a twilight interval lasting for several hours, which reminds one of the recovery from ether anesthesia or the sobering up after intoxication. A headache of varying severity is generally complained of after the attack, often lasting for several hours, and the patient is frequently drowsy and sleepy for some time. Mental depression following the attacks recalls strongly that which occurs after epileptic seizures. It is a common aftermath.

Case 5.—A woman, aged 24, a teacher, while attending a short course of instruction in a normal school near Baltimore was brought unconscious to the hospital with a history of having fallen over in a faint in the classroom about two hours previously. When seen she showed no evidence of trauma as a result of her fall. The temperature was subnormal, 97.8 F. by rectum. The pupils were widely dilated, the neck was slightly stiff, the breathing slow and stertorous, the skin moist and the face and forehead perspiring freely. The limbs were equally flaccid, and the deep reflexes were absent or obtained with great difficulty. The heart and lungs were normal, and the blood pressure was 110 systolic and 60 diastolic. The urine was free from sugar, albumin and casts. The girl's previous medical history was not at once obtainable, as she had arrived in Baltimore two days previously. Poisoning was considered a possibility, and gastric lavage was performed, as a result of which only a large amount of undigested food was recovered. The complete and mystifying absence of any indication as to the cause of the continued unconsciousness after two hours' observation made a lumbar puncture appear necessary, when the girl suddenly awoke, sat up and asked for water. She then said that she had been under treatment for diabetes mellitus in a middle western city, had ceased visiting her physician about three weeks before, but had continued to take insulin. On the morning of admission, feeling hungry, she had taken a larger breakfast than usual and a double dose of insulin. She had felt rather faint in the class-room and then had known nothing more until she opened her eyes in the hospital. When her history was thus related, a belated specimen of blood was taken for an estimation of sugar, and the blood sugar value was reported as 65 mg. per hundred cubic centimeters. For the rest of the day following her recovery she suffered from a severe occipital headache.

Case 6.—A woman, aged 20, who had been under treatment for diabetes of maximal severity in the clinic for upward of three years, and who took from 40 to 60 units of insulin a day, was brought into the hospital comatose at 2 a. m. Neighbors in an adjoining house had been awakened by screams and cries, had forced their way into her room, and had found her lying on the floor. A blood

sugar estimation showed that there were 50 mg. per hundred cubic centimeters. She was at once given dextrose intravenously and by rectum, but notwithstanding the almost immediate return to a normal or even high blood sugar level, and the appearance of glycosuria, she did not recover consciousness. She cried at intervals during the following day, resisted violently all efforts at treatment or feeding, and was totally unresponsive to any questioning. Most of the time she lay with eyes half closed, staring at the ceiling as if in a dazed condition. Gastric lavage on admission was returned with a large amount of undigested food. As she had previously been despondent over her condition, the possibility of poisoning was also considered, but analysis of the gastric contents revealed nothing abnormal, and the results of examination of the blood for carbon monoxide were negative. On the second morning, about sixty hours after the beginning of the attack, she awoke, conscious and completely oriented, but with no recollection of the previous events. On the night previous to admission she had taken her usual evening dose of insulin at 9 o'clock when she ate her supper, and had gone to bed. About an hour later, she had awakened with a sense of oppression in the abdomen and a feeling of weakness and hunger. Recognizing the symptoms, she had tried to get out of bed for food, but had collapsed on the floor. She knew nothing more of what had occurred. During the entire time from admission to the hospital until the recovery of consciousness sixty hours later, sugar was constantly present in the urine, and the blood sugar ranged from 120 to 180 mg. per hundred cubic centimeters.

CASE 7.—A boy, aged 16, with diabetes of maximum severity, received four injections of insulin a day during a course of treatment in the hospital: 20 units before breakfast, 15 units before lunch, 20 units at 5 o'clock and 15 units at 10 p. m. This dosage and the distribution of the daily diet into four portions following each of the injections of insulin finally rendered the urine sugar-free, and estimations on the blood sugar before each of the meals were between 110 and 160 mg. per hundred cubic centimeters. The patient felt well on this regimen, which was followed without incident for a period of a week. One night he complained of slight epigastric pain and had a loose bowel movement just before supper, but the insulin and supper were given as usual, as was the late evening dose and the late lunch following it. Next morning at 6 o'clock the nurse on making her morning rounds was unable to awaken the patient. On examination shortly after, he was found to have a subnormal temperature, and it was impossible to arouse him. There were slight twitchings of the face, and about the mouth; the extremities were relaxed and the deep reflexes were sluggish. The breathing was slow and deep, about 14 respirations a minute. The pulse rate was 56. The bladder was moderately distended. A blood sugar determination made at once showed 85 mg. per hundred cubic centimeters, and a catherterized specimen of urine was free from sugar or acid bodies. As no other explanation seemed adequate to account for his condition, and notwithstanding the fact that the blood sugar was little below the normal fasting level, an intravenous injection of dextrose was given, whereupon the patient became conscious and normally oriented within ten or fifteen minutes. He was totally unaware of what had happened since receiving the injection of insulin and lunch about eight hours previously.

Wilder [16] has recently stated that an increase in blood pressure occurs during insulin reactions as a constant and hitherto unmentioned phenomenon. In some of our cases a rise of from 20 to 30 mm. in systolic pressure has occurred, with somewhat less increase in the diastolic pressure, so that the pulse pressure was increased. When the reaction is severe, however, or when complete or partial lack of con-

16. Wilder, Russell M.: Hyperthyroidism, Myxedema and Diabetes, Arch. Int. Med. **38**:736 (Dec.) 1926.

sciousness supervenes, we have not found the pressure raised, and in the cases of actual coma it may be low. This is of some importance in differential diagnosis in cases in which patients are brought in unconscious, and in which in the absence of a history or of obvious marks of hypodermic injections, as has happened in three of our cases, the nature of the condition is doubtful. In these cases dilated pupils were present and the eyegrounds did not present abnormalities. The urine was free from albumin, and, as was to be expected, from sugar.

The diagnosis between a condition resulting from overdosage with insulin and diabetic coma does not usually present great difficulties even without laboratory aids. In the one condition, slow, shallow and at times irregular breathing occurs; in the other, hyperpnea, while the urinalysis quickly reveals the true state. Nevertheless, the tendency in some cases of diabetic coma toward a diminution of the urinary concentration and excretion of sugar is well known; and although none of the patients that we have observed have been free from glycosuria, such a condition has been reported in the literature. Patients in diabetic coma almost invariably show evidences of considerable or extreme dehydration, and such is not usually the case in coma due to insulin. The urine from our patients with severe insulin reactions has never shown the presence of acetone bodies. On the other hand, cases of diabetic coma have been reported in which acidosis has been due, at least in large part, to acids other than the acetone bodies. A determination of the blood sugar concentration and plasma bicarbonate will usually reveal the true condition and the presence or absence of acidosis.

Sevringhaus in reporting recently five cases with marked mental disturbance and unconsciousness lays stress on the production of these symptoms by repeated overdosage with insulin. Although severe reactions as a rule occur in patients with severe diabetes when large doses of insulin have been given over considerable periods of time, and while it would appear that, in general, emaciated persons are more prone to toxic reactions than are the well nourished, these generalizations are subject to exceptions. Insulin reactions of all types of severity may occur at any age, and mental disturbances in our series have not appeared more frequently in those who have previously had milder reactions.

It seems clear that the state of hypoglycemia alone is not always the cause or even the most important factor in the production of toxic symptoms. Patients in whom unconsciousness occurs may not revive for hours after the blood sugar has been elevated to normal or even higher by the administration of carbohydrate. On the other hand, recovery of complete consciousness may occur spontaneously even when the blood sugar is at a low level. Reactions of all shades of severity occur at various levels of the blood sugar. The series of cases recently reported by John[6] is of interest in this regard. Of twenty-four cases

studied, a blood sugar concentration of 80 mg. per hundred cubic centimeters or less occurred at the time of the reaction in only thirteen, while in the remaining eleven, it was above 80 mg. per hundred cubic centimeters, and in five of these patients it was actually over 200 mg. per hundred cubic centimeters when symptoms occurred. We have never seen such extreme cases of hyperglycemia in association with insulin reactions. We have, however, seen characterstic toxic insulin effects in four persons in whom the blood sugar taken at once and previous to the administration of carbohydrate was between 150 and 110 mg. per hundred cubic centimeters. The following case is typical of this group:

Case 8.—A diabetic patient, aged 58, who had never received insulin, and whose fasting blood sugar was 340 mg. per hundred cubic centimeters, was given an injection of 20 units of insulin during some metabolic studies. In the course of twenty minutes he became uneasy, faint and hungry; he sweated profusely, became pale and his blood pressure fell from 160 mm. systolic and 95 mm. diastolic to 130 mm. systolic and 80 mm. diastolic. A specimen of blood for the estimation of sugar was taken, and it was found to be 150 mg. per hundred cubic mentimeters. The urine was free from sugar and acetone bodies. As the patient's uneasiness grew much worse, the studies undertaken were discontinued, and the patient was given orange juice with cane sugar, which produced prompt abatement of all symptoms.

SUMMARY

The cases which have been studied indicate plainly the fact that unconsciousness due to insulin overdosage may supervene with great rapidity, often without warning in sleep, and with few or negligible prodromes. They also indicate that there is not always a set progression of symptoms such as is usually described. Not always do subjective symptoms of uneasiness or weakness or hunger precede more pronounced symptoms of pallor or flushing or sweating, in their turn preceding convulsions and unconsciousness. Generalized convulsions are exceptional in adults, although muscular twitchings, especially of the facial muscles and of the muscles of the extremities, are common. The clinical picture in two of our cases strongly suggested severe carbon monoxide poisoning, except that the coloring was not typical. The administration of carbohydrate to unconscious persons in insulin reactions does not always have the immediate prompt effect that we have been led to anticipate. It seems clear that the degree of hypoglycemia does not accurately determine the severity of the reaction, nor does the administration of sugar always relieve it. It is probable that the toxic effects may be due partly, or in certain cases entirely, to some other action of insulin. As Labbé has suggested, it is not possible to transfer directly to man the results of overdosage with insulin in animals. The psychic manifestations which are so common in man do not usually occur in animals, and in man there is wide individual variation in the response to overdosage.

THE SPECTROPHOTOMETRIC DETERMINATION OF HEMOGLOBIN *

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Within the last few years, a number of workers in the field of medicine have expressed dissatisfaction with the methods commonly used in determining the amount of hemoglobin in the blood. They have pointed out not only that the different types of hemoglobinometer use different values of concentration of hemoglobin for 100 per cent on the scale, but also that the methods themselves are for the most part not sufficiently accurate for use in diagnosis. Of these two objectionable features, the latter is, of course, by far the more serious. The confusion and misunderstanding caused by dissimilar scale values can be avoided simply by the adoption of a uniform method of reporting hemoglobin, as recommended recently by Lindsay, Rice and Selinger;[1] but if the method itself is not sufficiently accurate, errors in diagnosis are unavoidable.

ACCURACY OF METHODS IN COMMON USE

In order to obtain some idea concerning the accuracy of the more commonly used methods for estimating hemoglobin values, it is pertinent to the subject under discussion to examine certain data which have recently appeared in medical journals.

In table 1 are shown the results of comparative readings made by various observers. All values have been reduced to the same scale for comparison, so that the various scale standards used in the different methods do not affect the results here shown. The readings obtained by Senty[2] in his studies are indicated by points placed on several graphs exhibited in his paper. We have computed the average and maximal percentage differences from the values thus indicated by Senty. The same is true of the readings obtained by Brown and Roth,[3] and we have computed the percentage differences shown in table 1.

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1. Lindsay, J. W.; Rice, E. C., and Selinger, M. A.: A Plea for a Standardized Method of Estimating and Reporting Hemoglobin Values, J. Lab. & Clin. Med. **11**:737, 1926.

2. Senty, E. G.: A Comparative Study of Various Methods of Hemoglobin Determinations, J. Lab. & Clin. Med. **8**:591, 1922-1923.

3. Brown, G. E., and Roth, G. M.: Prognostic Value of Anemia in Chronic Glomerular Nephritis, J. A. M. A. **81**:1948 (Dec. 8) 1923.

It is evident from these results that the hemoglobin values given by the various methods differ greatly. When one considers that, with only two exceptions, no two methods here compared are found to agree on the average even to within 10 per cent, and that the maximal differences run from 26 to 56 per cent, one is convinced that altogether too much confidence is commonly placed in the readings given by the ordinary routine methods here represented.[4] These data do not give conclusive evidence concerning the relative accuracy of the different methods, since only the last three sets of readings in the table are compared with a method of proved accuracy (the van Slyke). The following points are worth noting, however. Readings by the Newcomer method are found by Lindsay, Rich and Selinger to vary from the van Slyke readings by

TABLE 1.—*Comparative Readings of Hemoglobin by Various Methods*

			Difference (per Cent) From Result by Method Indicated Below			
			Cohen and Smith		Newcomer	
Observer	Method	Observations	Average	Maximal	Average	Maximal
Senty	Dare	91	17.3	40	13.9	29
	Newcomer	91	6.0	40		
	Tallqvist	91	17.2	56	14.8	31
			Haldane-Palmer			
Brown and Roth	Dare	72 (0-70 per cent on Dare scale)	10.6	26		
		89 (above 70 per cent on Dare scale)	27.5	46		
			Van Slyke Oxygen Capacity			
Lindsay, Rice and Selinger	Dare	(3 instruments) 7	23.7	Not given by authors		
		3	27.3			
		2	38.0			
	Newcomer	7	2.1			
	Sahli	7	36.9			

only 2.1 per cent. However, too few comparative readings (seventeen) were taken to warrant conclusions. Senty finds the Newcomer method to give readings differing on the average by only 6 per cent from the Cohen and Smith readings; yet many of the individual differences are great, varying from —24 per cent (not shown in table) to +40 per cent. Only 82 per cent of all the ninety-one readings are in agreement to within 10 per cent. Therefore, judging from these data, we must conclude that neither the Newcomer method nor the method of Cohen and Smith is shown to be highly reliable, although the evidence is somewhat favorable to them.

All the data concerning the Dare hemoglobinometer lead inevitably to the conclusion that the readings are unreliable, unless one is willing to

4. The van Slyke oxygen-capacity method is sufficiently accurate, but it is not ordinarily used as a routine procedure because of the labor and time involved.

attribute the lack of agreement to the other four methods with which the Dare is compared. The latter alternative is not reasonable, however, especially since one of the four methods, the van Slyke, is known to be accurate to within 1 or 2 per cent. Again, it is interesting to note that Senty finds the Dare readings to agree with the Cohen and Smith and with the Newcomer readings little better than do the readings by the Tallqvist method. Since the latter method is commonly, and correctly, regarded as unreliable, this comparison is further evidence of the inaccuracy of the Dare method.

As to the Sahli hemometer, the seven readings taken by Lindsay, Rice and Selinger indicate that the instrument is highly inaccurate. Possibly the average difference from the van Slyke readings would be lower if more readings were compared. It is clear, however, that no method can be considered reliable in diagnosis if seven values are found to show an average error of 36.9 per cent.

We are hardly justified in drawing conclusions as to the accuracy of the Haldane-Palmer method, since it is compared only with the Dare method, with which it is in poor agreement.

Observations on the data in table 1 may be summarized briefly in the following conclusions:

1. All the evidence here shown indicates that the Dare, the Sahli and the Tallqvist methods are entirely too unreliable for use in accurate diagnosis.

2. The evidence concerning the method of Newcomer and that of Cohen and Smith is partly favorable and partly unfavorable.

3. No reliable conclusion can be drawn from these data concerning the Haldane-Palmer method.

OBJECT OF THE INVESTIGATIONS

The object of the present work has been to investigate the possibilities of the spectrophotometric method of determination of hemoglobin: (1) for the use in routine diagnosis, (2) for standardization purposes, and (3) as a research method. Briefly stated, the investigations were prompted by: (1) a growing dissatisfaction with the methods now in common use, because of their general unreliability, as illustrated in table 1; (2) the need for a standardizing method which is at once highly accurate, fairly simple, and capable of being reproduced at any time without uncertainty or ambiguity, and (3) the desirability for research workers to have at their disposal a method sufficiently simple and accurate to be used in following closely the course of treatment in its effect on hemoglobin concentration, or in investigating any question which may arise in regard to the value or variation of the concentration in states of health and disease.

THEORY AND EXPERIMENTAL METHOD

The theory of spectrophotometric determination of the amount of hemoglobin or other substance in solution is well known. When light is allowed to pass through a solution of a colored substance, such as hemoglobin, it is found in general that the different wave lengths are transmitted unequally. If one plots the fraction of light transmitted at each wave length, the spectral transmission curve for that particular substance is obtained. No two substances have spectral transmission curves of exactly the same shape; so in general one may with certainty identify a substance from the shape of its curve. The transmission of light of various wave lengths (from red to violet) by oxyhemoglobin (curve 1) and acid hematin (curve 2) are given in chart 1. For curve 1 the dilution was one part by volume of blood to 200 cc. (approximately) of 0.1 per cent solution of sodium carbonate; in curve 2 the dilution was also 1:200.

Again, the fraction of light transmitted by the substance at any given wave length can be used in calculating the concentration of the substance in the solution, providing the value of a certain constant called the absorption ratio is known for that wave length. The value of this absorption ratio is given by the relation

$$A = CD' \qquad (1)$$

in which A is the absorption ratio, C is the concentration of the substance and D' is the thickness of solution which transmits only one tenth of the amount of light (of that particular wave length) which enters it. From this relation, one has

$$C = \frac{A}{D'} \qquad (2)$$

As a rule, however, it is not convenient to make the thickness of solution examined exactly equal to the length D'. If the thickness is some other length, D, then it may easily be shown that

$$D' = -\frac{D}{\log_{10} F} \qquad (3)$$

in which F is the fraction of light transmitted by the thickness D. Substituting this value in equation (2), one has

$$C = -\frac{A}{D}\log_{10} F \qquad (4)$$

Since A and D are constants, this equation simply means that the concentration of the hemoglobin or other substance in the solution is proportional to the negative logarithm of the fraction of light transmitted.

This gives a simple method for estimating the amount of hemoglobin in blood. The values of A and D being known, it is necessary only to measure the fraction of light transmitted and to substitute this number for F in equation (4); or, simpler still, it is possible to make use of a curve plotted from equation (4) to read the concentration of hemoglobin directly from the curve as soon as the value of F has been found. A curve of this type, used in our investigations, is shown in chart 2. If the answer is obtained from a curve in this manner, and if one uses a direct reading spectrophotometer in finding F, then no computation of any kind is required.

As a rule, the spectral transmission curve of a colored substance in solution shows one or more absorption bands. The number, position and intensity of these bands are characteristic of the absorbing substance, differing in general even for different derivatives of the same substance. In the present determinations of concentration of hemoglobin, the transmission in the region of maximal absorption in one of the absorption bands of oxyhemoglobin was measured. The band chosen was the so-called β-band situated in the greenish yellow region of the spectrum. In this band, maximal absorption takes place at a wave length of approximately 542 millimicrons. This was determined by a series of examinations

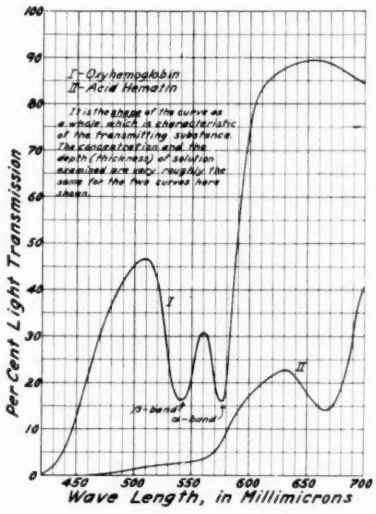

Chart 1.—Characteristic spectral transmission curves of oxyhemoglobin and acid-hematin. I is the curve for oxyhemoglobin; II, the curve for acid-hematin. It is the shape of the curve as a whole which is characteristic of the transmitting substance. The concentration and the depth (thickness) of solution examined are roughly the same for the two curves.

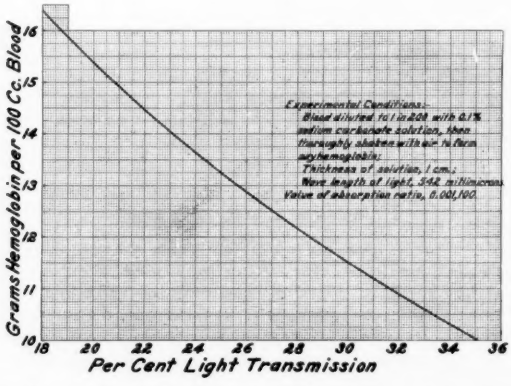

Chart 2.—Curve from which the hemoglobin concentration was read off without computation. Experimental conditions were: blood diluted to 1:200 with 0.1 per cent sodium carbonate solution, then thoroughly shaken with air to form oxyhemoglobin; thickness of solution, 1 cm; wave length of light, 542 millimicrons; value of absorption ratio, 0.001,100.

of the shape of the transmission curve in this region. Accordingly all measurements were made at a wave length of 542 millimicrons.

All transmission readings were taken on the direct reading spectrophotometer of recent design, manufactured by Keuffel and Esser, and called by them a "color analyzer." This instrument is more conveniently and rapidly operated than the usual type of spectrophotometer. To obtain light of any particular wave length, it is necessary only to turn a graduated dial to the proper setting. The light comes from two brightly illuminated blocks of magnesium carbonate situated at the rear of a lamp-house containing two 400 watt lamps, and passes through two parallel tubes with glass ends, one containing a solution of the substance to be examined, the other containing the solvent only. Equal illumination of the two halves of the optical field is obtained by rotating a graduated cylindric head by which the amount of light of any desired wave length passing through the standard solvent may be modified at will, and from which the percentage of transmission may be read off directly. Thus the fraction of light transmitted at any given wave length may be determined without computation of any kind.

The following technic was used in the determinations of hemoglobin concentration. The specimen of blood was obtained by venipuncture and at once shaken with about 20 mg. of powdered potassium oxalate to prevent clotting. As a rule, about 20 cc. of blood was drawn in order to insure a sufficient quantity for the various tests which it might be desirable to make. However, only a small amount was necessary for use in the hemoglobin determination. A small part (usually 0.05 cc.) of the specimen of blood was diluted to 1:200 with 0.1 per cent sodium carbonate solution, by means of carefully calibrated pipets. It was then thoroughly shaken with air to change the hemoglobin to oxyhemoglobin; one of the two analyzer tubes of the spectrophotometer was filled with the solution, and the fraction of light transmitted at a wave length of 542 millimicrons was obtained by averaging several (usually ten) readings. Analyzer tubes of 1 cm. in length were used. The fraction of light transmitted gave the value of the term F in equation (4), so that the hemoglobin concentration, C, could be calculated. Instead of substituting in equation (4), however, our results were read off directly from a curve (chart 2) obtained by plotting equation (4) as previously described.

The procedure therefore consists of the following steps: (1) dilution of the specimen of blood, (2) shaking with air, (3) filling the analyzer tube with standardized solution, (4) taking the transmission readings, and (5) reading the hemoglobin concentration from the curve. If the dilution is made with a diluting pipet similar to those used in blood counting, the entire procedure can be carried out in about five minutes. The method therefore is neither laborious nor time consuming. There are no corrections to be made. Computations of any kind are not necessary, since the result is obtained from a curve.

DETERMINATION OF THE ABSORPTION RATIO

In order to use equation (4) it was necessary to know the value of the absorption ratio A for oxyhemoglobin for the wave length employed, namely, 542 millimicrons. Williamson,[5] in his studies on hemoglobin in 1916, found the average value of A for oxyhemoglobin for the spectral region between wave lengths 534 and 542 millimicrons to be about 0.00118 (the average of two determinations, 0.001168 and 0.001190). The value at wave length 542 millimicrons has never before been determined, however. In determining it the following method was employed.

Fifteen different specimens of blood, obtained by venipuncture and treated with a small amount of powdered potassium oxalate to prevent clotting, were tested for hemoglobin content by the van Slyke oxygen capacity method. For these tests we are indebted to Dr. E. J. Baldes, of the Section on Physics of the

5. Williamson, C. S.: Influence of Age and Sex on Hemoglobin, Arch. Int. Med. **18**:505 (Oct.) 1916.

Mayo Clinic. The tests were carefully carried out, with the latest improved van Slyke apparatus. In every case (except one) two or more independent determinations were made and the average taken as the correct value of the hemoglobin concentration for the specimen under test. It is felt, therefore, that considerable confidence can be placed in the accuracy of the results. All the specimens were tested for methemoglobin by a sensitive spectrometer, and it was not found in any. Such tests were thought desirable since the presence of methemoglobin would affect the comparative values as determined by the oxygen capacity and spectrophotometric methods. Part of each specimen of blood was also diluted to 1:200 with 0.1 per cent sodium carbonate solution, shaken thoroughly with air to secure oxygenation, and tested with the direct reading spectrophotometer for transmission at a wave length of 542 millimicrons, the procedure being exactly the same as that followed in obtaining the hemoglobin concentration by the spectrophotometric method. In order to increase the accuracy, two or three (three in all but two instances) different dilutions of each specimen of blood were made up and examined. Ten settings of the instrument were made for each dilution. The average value of the transmission for the two or three dilutions was then substituted for F in equation (4), the concentration C being the

TABLE 2.—*Absorption Ratio for Oxyhemoglobin at Wave Length 542 Millimicrons*

Determination	Absorption Ratio A
1	0.001088
2	1088
3	1097
4	1095
5	1088
6	1090
7	1083
8	1111
9	1080
10	1124
11	1113
12	1103
13	1132
14	1108
15	1095
Average	0.001,100 ± 0.000,003

value found by the van Slyke method. The value of D was 1, since containing-tubes 1 cm. in length were used. The value of the absorption ratio A was then found from the equation.

The values obtained for A, the absorption ratio, are shown in table 2. Since fifteen independent determinations were made, and since the probable error is about 3 parts in 1,100, it is believed that the average value of A equals 0.001100 is fairly accurate. It was used as the absorption ratio for oxyhemoglobin at wave length 542 millimicrons in all the measurements of hemoglobin concentration reported in this paper. Once established, the value of this ratio is, of course, a constant, which can be used in all measurements at the wave length, whatever type of spectrophotometer is used.

TESTS OF ACCURACY OF SPECTROPHOTOMETRIC, DARE AND ACID-HEMATIN COLORIMETRIC METHODS

When the value of A had been determined, the hemoglobin concentration for each of the fifteen specimens of blood used in determining A was obtained from a curve, as previously described. These values were compared with the van Slyke readings in order to obtain a check on the accuracy of the spectrophotometric method. At the same time, each specimen was examined by the acid-hematin colorimetric (Cohen and Smith) and Dare methods in order to obtain first-hand information as to their accuracy. A colorimeter of the Duboscq type was used in

the acid-hematin method. Readings on the Dare instrument were made by two different observers, both experienced in colorimetric readings. Each made six settings on the same instrument under identical conditions. The average of each set of six readings was used in obtaining the concentration. Results by all three methods are shown in table 3 and are expressed in percentage variations from the van Slyke results. The values shown are not affected by the scale standards used by the acid-hematin and Dare methods, since all readings were converted to grams of hemoglobin for each 100 cc. of blood before comparisons were made.

DISCUSSION OF RESULTS

In order to determine correctly the accuracy of any method of measurement, one must know whether any of the values obtained in the series of measurements were arbitrarily discarded. It is obvious that if the experimenter discards the results which differ widely from the correct value and reports only those which are more accurate, then a correct

TABLE 3.—*Percentage Variations from van Slyke Readings*

Specimen of Blood	Spectrophotometer			Acid Hematin (Cohen and Smith)	Dare	
	Dilution No. 1	Dilution No. 2	Dilution No. 3		First Observer	Second Observer
1	1.7	1.3	0.3	2.3	13.6	6.3
2	1.8	1.3	0.2	4.6	15.6	1.9
3	0.7	0.1	...	6.0	20.1	2.3
4	0.5	1.5	...	1.3	13.6	5.8
5	1.8	1.1	0.4	1.2	26.1	16.5
6	1.5	0.7	0.9	0.4	26.7	11.2
7	1.6	1.9	1.2	1.2	23.5	20.2
8	0.6	0.8	1.6	1.0	11.7	3.5
9	2.3	1.3	1.9	1.4	13.7	0.1
10	2.1	2.7	1.8	1.8	21.6	14.6
11	1.6	0.1	1.8	2.7	24.7	12.7
12	0.0	0.2	0.9	3.3	14.5	7.6
13	3.3	2.4	2.7	1.4	10.7	4.9
14	3.0	0.0	0.9	7.7	17.8	10.9
15	1.1	0.6	0.4	0.5	0.8	12.9
Average		1.3		2.5	17.0	8.8
Maximum		3.3		7.7	26.7	20.2

idea of the accuracy of the method cannot be obtained from the results published. Again, it is evidently of great importance to the diagnostician to know approximately the maximal error to which the measurements are liable, since that maximal error determines the limit of reliability of the diagnosis in any particular case.

With these principles in mind, it is possible to evaluate the results shown in table 3. No results were arbitrarily discarded in making up the table; that is, the results shown in the table represent an unbroken series of fifteen measurements (actually forty-three measurements by the spectrophotometric method and thirty by the Dare), all results in the series being included regardless of their degree of accuracy.[6] One may, therefore, take the series as a fairly reliable criterion of the dependability of the methods represented.

6. Results in one case were thrown out, but only because the values given by the van Slyke method were not consistent.

From these results the high degree of accuracy of the spectrophotometric measurements is apparent. For all the forty-three independent determinations, the average variation from the results by the van Slyke method is only 1.3 per cent, with a maximal variation of 3.3 per cent. As the individual determinations by the van Slyke method itself may frequently be in error by 1 per cent or more, according to careful check by other experimenters, this agreement must be considered to be close. The maximal error observed probably represents the sum of two errors, that of the van Slyke method and that of the spectrophotometric procedure. The average error should not be affected by the inaccuracies of the van Slyke results, however, since they are probably as often positive as negative. It may be concluded, therefore, that under reasonably satisfactory conditions of measurement it should be possible to measure the concentration of hemoglobin in the blood by the spectrophotometric method with an average error of 1.5 per cent or less, and with a maximal error of about 3 per cent.

Results obtained by the acid-hematin colorimetric method also were found to agree well with the van Slyke results, with an average variation of 2.5 per cent and a maximal variation of 7.7 per cent. The errors to be expected, therefore, are roughly twice those indicated for the spectrophotometric method. It is to be noted that the standard solution here used in the determinations for acid hematin was calibrated from the van Slyke readings on the fifteen specimens of blood, so that any inaccuracy of calibration of the standard solution did not affect the accuracy of the results here indicated. The degree of accuracy here shown can therefore be expected only in case the standard solution is accurately calibrated at the time the readings are taken.

The present results agree roughly with those of other experimenters in regard to the inaccuracy of the Dare method (compare with table 1). Not only are the average and maximal errors high, but the two observers do not read the same instrument even approximately the same under identical conditions. In fairness to the method, it should be noted here that in nearly every case the Dare readings were too low. This in itself might indicate only that the instrument used needed recalibration. But inspection of the readings from which table 3 is made up, shows that the errors run from +12.9 per cent to —26.7 per cent. Therefore by recalibration of the instrument it is obviously impossible to reduce the inaccuracy of the method to satisfactory figures. It is clear that if experienced observers do not read the same instrument alike, and if an individual determination, which is the average of six settings of the instrument, may be in error by as much as 20 per cent or more, the instrument is not sufficiently accurate for careful work in diagnosis.

EVALUATION OF THE SPECTROPHOTOMETRIC METHOD

The accuracy of the spectrophotometric method of determining hemoglobin, judging from our results, appears to be such as to recommend it for practically all purposes. As used in the present investigations, it is also sufficiently simple and rapid to warrant its use in routine measurements. It should prove to be a convenient and accurate method for use in medical research and in the standardization of various instruments and methods.

SIMPLIFICATION OF THE SPECTROPHOTOMETER

The form of spectrophotometer used in the present work is open to the objection that it is expensive. It therefore became our desire to perfect a more simple and inexpensive instrument. For this purpose we chose a Wratten light filter (no. 74, Eastman Kodak Company) which permitted the passage of light of only a reasonably narrow region (from 510 to 550 millimicrons, with its maximum at about 530 millimicrons) covering most of the β-absorption band of oxyhemoglobin. The light passing through this filter is not monochromatic, but since it contains those wave lengths which are absorbed in one of the characteristic absorption bands of oxyhemoglobin, it may well be used in spectrophotometric measurements of hemoglobin concentration. By the use of this filter and a neutral density wedge for balancing intensities, we were able to construct, in the rough, an instrument which will cost little, and which will enable determinations of hemoglobin to be made rapidly and with a fair degree of accuracy. This instrument and some other simple designs are now under investigation.

In this connection, it may be noted that Kennedy[7] has recently described a method of using light filters in connection with a Duboscq colorimeter in hemoglobin measurements. Also, Exton[8] presented the results of some similar simplified spectrophotometric methods at the meeting of the Optical Society of America held in Philadelphia, October, 1926.

SUMMARY

Data obtained by various investigators clearly indicate that the Dare, the Sahli and the Tallqvist methods for the determination of hemoglobin are entirely too unreliable for use in making accurate determinations. The evidence concerning the method of Newcomer and that of Cohen and Smith is partly favorable and partly unfavorable.

The object of the present work has been to investigate the possibilities of the spectrophotometric method.

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7. Kennedy, R. P.: The Use of Light Filters in Colorimetry with a Method for the Estimation of Hemoglobin, Am. J. Physiol. **78**:56, 1926.

8. Exton, W. G.: A New Method of Colorimetry, J. Opt. Soc. Am. **14**:134, 1927.

In each of fifteen different specimens of blood the hemoglobin concentration was determined by four different methods; namely, the van Slyke oxygen capacity, the spectrophotometric, the acid-hematin colorimetric (method of Cohen and Smith) and the Dare. In the spectrophotometric method the hemoglobin was converted into oxyhemoglobin and transmission readings were taken at wave length 542 millimicrons. A direct reading spectrophotometer was used, and results were obtained from a curve without computation. It was first necessary, however, to determine the absorption ratio A for oxyhemoglobin for a wave length of 542 millimicrons. The value of A, the absorption ratio, was found to be $0.001100 \pm 0.000{,}003$.

Forty-three independent determinations by the spectrophotometric method (no results discarded) gave values having an average variation of only 1.3 per cent from the van Slyke values, with a maximal variation of 3.3 per cent. The method, therefore, is shown to be highly accurate.

Values given by the acid-hematin colorimetric method also agreed fairly well with the van Slyke values, differing on the average by 2.5 per cent, with a maximal difference of 7.7 per cent.

The Dare method was highly inaccurate. This agrees with results by other experimenters. Readings by two different observers differed from the van Slyke readings by 8.8 and 17 per cent on the average, with maximal differences of 20.2 and 26.7 per cent.

We conclude from our investigations that the spectrophotometric method of estimating hemoglobin is not only highly accurate, but also simple and fairly rapid as well. It should prove useful in diagnosis, in work of standardization and in research.

A simple and inexpensive form of spectrophotometer, using a color filter and a neutral density wedge, has been tried out and found capable of giving fairly accurate results.

EFFECT OF EXERCISE ON RESPIRATORY EXCHANGE IN HEART DISEASE. II*

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Although breathlessness on exertion is one of the most common symptoms of disease, and though difficulty in obtaining a sufficient exchange of oxygen and carbon dioxide is the underlying cause, the exact mechanism is little understood. Certainly it is the quantity of air and of oxygen relative to the capacity of the subject rather than the absolute amount which is important, and we have tried to learn more about this phenomenon by comparing the reaction of normal and of breathless subjects to the same exercise.

The pioneer work of Haldane has led to an enormous amount of investigation during the last twenty years, and has been applied to the changes at the beginning and end of exercise by Krogh and Lindhard,[1] by Campbell, Douglas and Hobson[2] and, more recently, by Hill and his fellow workers.[3] The changes at the beginning take place quickly, and the larger part of the reverse changes take place only a little less quickly when the exercise is stopped. It would be irrelevant to discuss this normal adaptation in detail in this article, but almost at the moment exercise is started, the pulse rate and the rate and depth of breathing are much increased. These take place through nervous influences at first, since, owing to the time taken by the blood in circulation, the chemical factors cannot come into play for some seconds and are not completely in operation for a minute or more, and until then the gaseous exchange gives no true indication of the actual metabolic changes. At the end of exercise all of these drop rapidly at first, then more gradually, and even after mild exercise the return is not complete for two or three minutes. The oxygen intake returns to normal rapidly, and the carbon dioxide output more slowly, so that at this period there is a high respiratory quotient.

Recent work on the blood and respiration in patients who are short of breath from heart disease has been summarized and discussed by

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*From the Medical Wards and the Department of Physiology, Guy's Hospital.

1. Krogh, A., and Lindhard, J.: J. Physiol. **47**:113, 1913; **53**:431, 1919.

2. Campbell, J. M. H.; Douglas, C. G., and Hobson, F. G.: Phil. Tr., Lond. B. **210**:372, 1920.

3. Hill, A. V.; Long, C. N. H., and Lupton, H.: Proc. Roy. Soc. Med. B. **96**:436, 475, 1924; ibid. B. **97**:84, 155, 1925.

Means [4] and by Meakins and Davies.[5] The most complete information could be obtained on the actual output of the heart per beat and its minute volume, but the different methods are not concordant or easy to use even in healthy subjects, though valuable results have been obtained. This method has been tried by Means and Newburgh,[6] and by Meakins, Dautrebande and Fetter [7] in valvular disease of the heart, the minute volume having been determined during exercise as well as during rest, and by Barcroft, Bock and Roughton in paroxysmal tachycardia.[8] Means and Newburgh examined a patient with aortic and mitral disease, whose heart was "well compensated"; with exercise there was less increase in the minute volume of his blood flow than normally. Meakins, Dautrebande and Fetter found that the minute volume even at rest tended to be less than normal and was much less when there was auricular fibrillation; with exercise most of the increase was obtained by increasing the pulse rate.

Few studies have been made on the respiratory exchange during and after exercise in patients with heart disease, but observations on young men with valvular disease have been made by Peabody and Sturgis.[9] The exercise used was running upstairs for one minute. These authors comment on the delayed fall of ventilation in patients with heart disease, on the delayed elimination of carbon dioxide with a consequent rise of quotient, on the lower percentage of carbon dioxide in the expired air, on the tendency to shallow breathing and increased total ventilation and on the fact that light exercise produced the same percentage of change in both groups, but more subjective dyspnea in the patients.

We have obtained a more detailed picture by measuring the respiratory exchange for each minute separately instead of for five minute periods, and by continuing the exercise for five or ten minutes instead of for one minute only, as earlier observations had shown that even three minute exercise was not long enough to obtain equilibrium.[10] The easier of our exercises was continued for ten minutes, but as the results showed that five minutes were really long enough to give the information required, the harder exercise was done only for this period. The tests chosen were stepping on and off a wooden block 13 inches high six or

4. Means, J. H.: Dyspnea, Baltimore, Williams & Wilkins, 1924, p. 79.

5. Meakins, J. C., and Davies, H. W.: Respiratory Function in Disease, Edinburgh, Oliver and Boyd, 1925, p. 295.

6. Means, J. H., and Newsburgh, L. H.: Tr. A. Am. Phys. **30**:51, 1915.

7. Meakins, J.; Dautrebande, L., and Fetter, W. J.: Heart **10**:153, 1922.

8. Barcroft, J.; Bock, A. V., and Roughton, F. J.: Heart **9**:7, 1921-1922.

9. Peabody, F. W., and Sturgis, C. C.: Clinical Studies on Respiration: Effect of Exercise on Metabolism, Heart Rate and Pulmonary Ventilation of Normal Subjects and Patients with Heart Disease, Arch. Int. Med. **29**:277 (March) 1922.

10. Campbell, J. M. H., and Poulton, E. P.: Quart. J. Med. **20**:27, 49, 1926.

twelve times a minute. At least three observations were made on each patient at both rates, except in one patient who performed the harder exercise.

The ventilation and metabolism were measured by a closed circuit apparatus of the Regnault-Reiset type developed with the help of Dr. E. P. Poulton [10] from similar methods used by Douglas and Haldane,[11] Krogh [12] and Benedict.[13] As we hope to publish a full description shortly, it need not be detailed in this article.

METHOD

Three patients short of breath as a result of heart disease were compared with a healthy medical student; only small numbers were examined, because a more accurate picture could be obtained by taking the average of several observations, and because in any respiratory work, breathing into a mask is always a disturbing factor which can best be eliminated by practice. These four subjects were examined frequently during a period of four months. Young patients with valvular disease of the heart were chosen because this had a relatively simple pathology, and other changes, such as renal or arterial disease, could be readily excluded.

CLINICAL CONDITION OF THE PATIENTS

It is difficult to choose for observation patients in whom there is one simple heart lesion such as might be produced in an experiment on animals. In two of these three cases the lesion was not simple, but this is perhaps less important than it seems at first, because, except for the different type of breathing found in aortic regurgitation and mitral stenosis, it appears to be the degree of the resulting disability rather than its nature which affects the respiratory exchange.

This disability is difficult to express exactly in words; none of these patients belonged to the class in which the cardiac lesion is of such slight importance as to interfere only with heavier work, or to the class in which the onset of cardiac failure has stopped all but the lightest exertion. They belonged to what is perhaps the commonest group of patients with valvular disease, outpatients, for whom walking is fairly easy, but for whom any other than the lightest occupation is impossible and the onset of definite cardiac failure a frequent threat. Two had already had attacks of cardiac failure from which they had made good, if only temporary, recoveries; while in the third, such an attack followed about nine months after these tests. At the time all were able to walk about slowly without much difficulty, but they could not do much more than this.

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11. Douglas, C. G., and Haldane, J. S.: J. Physiol. **44**:305, 1912.
12. Krogh, A.: The Respiratory Exchange in Animals and Man. New York, Longman, Greene & Co., 1916.
13. Benedict, F. G.: Deutsches Arch. f. klin. Med. **107**:156, 1912.

A more exact idea will be given by the result of the stepping tests. It should be remembered that, judged by the consumption of oxygen to which this exercise gives rise, it is a little easier to do six of these steps a minute than to walk two and one-half miles an hour; it is more difficult to do twelve steps a minute than to walk three and one-half miles an hour but easier than to walk four miles an hour, and it is more difficult to do eighteen steps a minute than to walk four and one-half miles an hour (Douglas [14]). Six steps a minute was easy for all these patients; twelve steps a minute was the first rate which appeared at all difficult. G. could not step faster than this, while the other two could start to do eighteen steps a minute but could not continue even for three minutes. The breathlessness seemed greatest in G. and least in L. A short clinical history of the three patients follows. Some preliminary results on the ventilation have been recorded in another paper,[15] in which L. was referred to as case 11, G. as case 12 and A. as case 17. The normal person was a medical student (F.J.S.), aged 24, who showed no clinical evidence of heart disease, and who had had no serious illness. He was fit and, though not in training, was taking a moderate amount of exercise.

The pulse rates and other results after these same stepping exercises have been recorded by Hunt and Pembrey.[16]

REPORT OF CASES

CASE 1.—L., a woman, aged 22, had rheumatic fever in 1913 and was treated in bed for seven weeks. When she left school in 1917, she was engaged in munition work and continued to feel well until 1921, when she again had rheumatic fever. Since then she had suffered from breathlessness and had been unable to work. She had recently coughed up some blood-streaked sputum. On admission to the hospital she was comfortable when at rest, but had a cough and was breathless on exertion. Her heart rate was irregular at the apex between 120 and 130, and at the wrist between 100 and 110. There was a diffuse impulse outside the nipple line, and the area of dulness extended almost to the anterior axillary line and to the right of the sternum. A systolic and a long diastolic bruit were heard at the apex. There were coarse râles at the base of both lungs, but no edema of the feet and no albuminuria.

Auricular fibrillation and mitral stenosis and regurgitation were diagnosed by Dr. Hunt. When digitalis was administered the pulse rate soon dropped, and her apex beat came in somewhat. The patient was allowed to be up after five weeks for graduated exercises and was discharged, able to walk about slowly without being short of breath and with an almost regular pulse rate of 70 when at rest. Her condition continued much the same; she attended the outpatient clinic regularly and took digitalis until these observations were made. There was little change in her general condition a year later.

CASE 2.—G. had rheumatic fever when he was 11 and was twice in the infirmary for eight and six weeks, respectively. He was still short of breath on exertion a year later when these tests were made. The area of dulness extended to the

14. Douglas, C. G.: J. Physiol. **42**:17, 1911.

15. Campbell, J. M. H.: Guy's Hosp. Rep. **76**:394, 1926.

16. Hunt, G. H., and Pembrey, M. S.: Guy's Hosp. Rep. **71**:415, 1921; **72**: 367, 1922.

right of the sternum and half an inch to the left of the midclavicular line. There was a presystolic bruit and thrill at the apex, and the pulse rate at rest was 100 and regular. He did not have edema of the feet. Nine months after these tests, the patient became increasingly short of breath and was admitted to Guy's Hospital under the care of Dr. Beddard, who diagnosed mitral stenosis and regurgitation with heart failure. His pulse was still regular, but the rate had increased to 130. He had edema of the feet, crepitations at the base of the lungs, albuminuria and an enlarged liver. The patient improved under the intravenous administration of strophanthin; but six months later, he was still just able to walk about and was in a much worse condition than when these tests were made.

Case 3.—A., a man, aged 21, gave no history of rheumatic fever. A year before admission to the hospital he had noticed breathlessness, which had gradually become worse. On admission he did not appear to suffer from breathlessness or cyanosis when at rest, but he became breathless on exertion. His pulse was irregular in rhythm and force (between 100 and 120), and some beats felt at the apex were not felt at the wrist. There was visible pulsation over a wide area in the epigastrium and as far as the left anterior axillary line, where the apex could easily be felt in the sixth intercostal space. The area of dulness extended nearly to this line and to the right of the sternum. There were diastolic and systolic bruits at the apex and in the aortic area. There was no edema of the feet and no albumin in the urine. The Wassermann reaction was negative.

Dr. French diagnosed auricular fibrillation, aortic stenosis and regurgitation, mitral stenosis and adherent pericardium. The patient improved on digitalis therapy and was able to get up after a fortnight. He was able gradually to get about the ward without raising his pulse rate above 80 or 90, and was discharged after a month. He attended the outpatient clinics regularly, and graduated exercises were prescribed, but he was not able to progress far with these or to return to work as a vanman, although he was anxious to do so. These tests were made about a year after his discharge from the hospital. His pulse rate was then about 60, and almost regular, under the influence of digitalis, and his apex beat was not displaced so far to the left. Aortic regurgitation seemed to be the outstanding valvular lesion. He died about a year later, but unfortunately the details were not known.

THE OXYGEN INTAKE DURING AND AFTER EXERCISE

The figures for each subject are given in tables 1 and 2, but to make any comparison, allowance must be made for the difference in weight. At rest, metabolism varies with surface area, but the extra metabolism due to exercise varies with the work done; this depends on the body weight, because the height of the step up which the body weight was lifted was the same. The best way to compute the oxygen intake, therefore, was to deduct the metabolism at rest from that during exercise and to express the difference as cubic centimeters per kilogram of body weight. The weight of the normal subject was 75 Kg. and of L., G. and A. 47.5, 23.8 and 48.2 Kg., respectively.

When this was done, there was surprisingly little difference in the oxygen intake. The normal subject used a little less than the others, especially during the harder exercise. The mechanical work done at both rates was the same, as the easier exercise was continued twice as long, but the extra metabolism for the easier exercise was greater because of the larger number of stepping movements. During the easier exercise, the extra oxygen intakes expressed as cubic centimeters per

TABLE 1.—*Metabolism Each Minute with Exercise (Six Steps a Minute)*

Period	Min.	Normal Subject (5)‡				L. (3)‡				G. (3)‡				A. (3)‡			
		Pulmonary Ventilation, Liters	Carbon Dioxide, per Cent	Carbon Dioxide, Cc.	Oxygen, Cc.	Pulmonary Ventilation, Liters	Carbon Dioxide, per Cent	Carbon Dioxide, Cc.	Oxygen, Cc.	Pulmonary Ventilation, Liters	Carbon Dioxide, per Cent	Carbon Dioxide, Cc.	Oxygen, Cc.	Pulmonary Ventilation, Liters	Carbon Dioxide, per Cent	Carbon Dioxide, Cc.	Oxygen, Cc.
At rest	..	6.9	3.39	234	267*	5.9	3.10	183	231*	5.1	2.55	131	167*	8.2	2.40	196	230*
During exercise	1	12.3	3.99	492	550	8.3	3.23	268	433	6.0	2.69	160	250	8.5	3.06	260	417
	2	13.9	....	580†	620	9.6	....	341†	483	8.0	....	210†	267	9.8	....	330†	517
	3	13.8	4.25	587	720	9.6	....	380	517	9.1	....	237	283	11.3	....	380	533
	4	14.0	....	598	760	10.1	....	398	533	10.4	2.41	254	300	12.8	3.40	435	550
	5	14.7	4.31	634	760	10.1	3.98	403	533	10.7	....	264	317	12.8	....	470	566
	6	14.5	....	620	750	10.4	....	414	533	11.6	....	270	340	13.9	3.57	497	566
	7	14.7	....	627	740	10.7	....	421	550	11.6	2.38	274	340	14.4	....	497	533
	8	15.2	4.24	645	744	10.7	4.00	428	550	12.0	....	276	340	14.4	....	510	550
	9	14.9	....	630	734	10.4	....	430	567	12.4	2.23	278	340	14.9	3.37	504	566
	10	15.2	4.21	640	734	10.9	3.96	432	567	13.0	....	280	340	15.7	....	520	550
At rest afterward	1	12.0	4.30	518	650	9.9	4.09	405	533	10.0	2.70	270	325	14.7	3.50	516	483
	2	9.5	3.69	351	370	9.3	3.76	350	366	9.6	2.48	238	250	11.2	3.25	364	300
	3	8.2	3.52	288	330	8.0	3.52	282	250	8.4	2.50	210	200	10.1	3.15	318	266
	4	7.5	3.48	262	320	7.7	....	240	266	7.2	2.49	184	200	9.3	3.07	286	250
	5	7.5	....	260	320	6.7	3.21	215	250	7.0	....	180	185	9.6	....	270	233
	6	7.3	3.45	252	310	6.0	....	198	217	6.7	....	168	185	9.1	2.79	254	233
	7	6.9	....	231	280	6.1	....	188	217	6.3	2.50	158	167	8.5	....	230	217
	8	7.3	3.43	249	290	5.9	3.10	183	233	5.6	....	148	167	8.3	2.63	219	233
	9	6.7	....	230	260	5.9	....	182	233	5.4	....	137	167	8.0	....	210	217
	10	6.6	....	226	270	5.9	....	181	233	5.2	2.53	132	167	8.0	2.58	207	217

* The average barometric pressure and room temperature to correct these figures to standard temperature and pressure are, respectively, 769 and 14 for the normal subject, 765 and 13.5 for L., 760 and 15.5 for G and 765 and 17.5 for A.

† When the carbon dioxide percentage was not determined, the carbon dioxide output was obtained by interpolation.

‡ The number of observations of which these figures are the average. The actual oxygen output would be given more accurately by the curves in figures 1 and 2, as the minute figures were recorded as the pointer passed each 50 cc. mark.

TABLE 2.—*Metabolism Each Minute During Exercise (Twelve Steps a Minute)*

Period	Min.	Normal Subject (3)‡				L. (3)‡				G. (1)‡				A. (3)‡			
		Pulmonary Ventilation, Liters	Carbon Dioxide, per Cent	Carbon Dioxide, Cc.	Oxygen, Cc.	Pulmonary Ventilation, Liters	Carbon Dioxide, per Cent	Carbon Dioxide, Cc.	Oxygen, Cc.	Pulmonary Ventilation, Liters	Carbon Dioxide, per Cent	Carbon Dioxide, Cc.	Oxygen, Cc.	Pulmonary Ventilation, Liters	Carbon Dioxide, per Cent	Carbon Dioxide, Cc.	Oxygen, Cc.
At rest	..	6.8	3.12	213	260*	6.0	3.13	188	230*	5.1	2.55	131	167*	8.3	2.42	202	232*
Exercise	1	13.2	4.27	562	650	10.5	3.63	381	450	7.2	2.20	158	250	11.5	2.87	330	500
	2	16.3	....	721†	900	12.9	....	496†	750	12.0	....	268†	350	13.1	....	422†	633
	3	17.9	....	808	900	13.5	4.02	539	750	14.4	2.28	328	400	15.3	3.25	492	700
	4	19.7	4.38	865	1020	14.0	4.05	568	783	....	....	...	...	17.6	....	572	750
	5	20.8	4.39	910	1010	14.8	4.03	600	750	....	....	...	...	19.0	3.32	632	780
At rest after exercise	1	16.2	4.09	662	730	12.4	4.20	542	783	12.8	2.49	320	400	17.5	3.32	592	645
	2	10.6	3.68	390	480	11.2	3.71	402	466	11.6	2.41	280	300	14.9	3.25	485	412
	3	8.9	3.57	318	380	9.7	3.64	342	366	10.4	2.51	262	250	12.5	3.22	402	333
	4	7.9	....	277	350	8.5	....	290	266	8.0	....	220	200	11.5	....	352	300
	5	7.2	3.55	256	290	7.7	....	260	250	6.4	....	190	200	11.2	2.94	330	288
	6	7.0	....	247	280	6.9	3.27	230	233	6.4	2.62	168	180	10.9	....	308	283
	7	7.0	3.39	238	290	6.4	....	220	225	5.6	....	146	170	10.1	....	278	275
	8	6.9	....	230	280	6.4	....	210	225	4.8	....	131	170	9.6	2.68	258	275
	9	6.9	....	225	250	5.9	3.13	196	225	4.8	2.55	122	160	9.6	....	252	267
	10	6.4	3.33	221	260	6.4	....	190	225	4.8	....	122	160	9.1	....	232	267
	11	6.3	....	219	260	5.9	....	187	233	....	....	...	...	8.8	2.52	220	250
	12	6.0	....	217	260	5.6	3.13	185	233	....	....	...	...	8.8	....	216	250
	13	6.2	....	215	260	6.1	....	184	233	....	....	...	...	8.5	....	214	250
	14	6.2	....	214	260	5.9	3.10	183	233	....	....	...	...	8.8	2.42	212	250
	15	6.2	....	213	260	6.1	....	182	233	....	....	...	...	8.8	....	210	250

* The average barometric pressure and room temperature to correct these figures for standard temperature and pressure are 757 and 15 for the normal subject, 757 and 16.5 for L., 763 and 16 for A. and 751 and 17.5 for G.

† When carbon dioxide percentage was not determined directly, the carbon dioxide output was obtained by interpolation.

‡ The number of observations of which these figures are the average. The actual oxygen output would be given more accurately by the curves in figures 1 and 2, as the minute figures were generally recorded as the pointer of the dial passed each 50 cc. mark.

kilogram were 70 for the normal subject against 73, 75 and 71 for the patients, and during the harder exercise, 58 for the normal subject against 69, 62 and 67 for L., G. and A., respectively. The differences were small, and the ability of these subjects to take exercise was but little influenced by the different amounts of oxygen they needed for the same work.

The proportion of this extra oxygen which had to be obtained after the exercise was finished showed a significant difference. At the more rapid rate the normal subject used only 13 cc., while the others used between 20 and 22 cc. per kilogram of body weight; in other words, only 23 per cent of the extra oxygen intake of the normal subject, but 30, 35 and 32 per cent of that of the three patients, took place after exercise. The slower rate of exercise was hardly sufficient to bring out this difference.

The relative oxygen intake for the patients can be compared with that for the normal subject in figures 1 and 2, which show the gaseous exchange expressed as cubic centimeters per kilogram. In the normal subject the oxygen intake rose rapidly and had practically reached its maximum by the third minute; indeed, when the exercise was continued for ten minutes, it fell somewhat. At the slower rate there was no significant difference in A.; in L., as much oxygen was obtained at the beginning of exercise but more was taken in at the end and for two minutes after, and in G. less was obtained at the beginning and more in the first four minutes after the exercise was stopped. At the faster rate the results were more striking, as would be expected from the total figures already given: in A., the oxygen intake was a little less at the beginning of exercise, but toward the end and for ten minutes after the extra consumption of oxygen, it was always slightly greater; in L. more oxygen was obtained at the end of exercise and considerably more in the first three minutes after, and in G. much less in the first two minutes of exercise and much more in the first four minutes after.

On the whole, there was a silghtly greater consumption of oxygen in the patients with heart disease, and a smaller proportion of the oxygen was taken during the exercise, especially during the harder one. While both these changes would tend to increase the breathing, the extent of the change was not sufficient to produce any degree of dyspnea normally.

In these patients it is interesting to compare the oxygen intake during exercise and the work done, as various observers have shown that there is a linear relationship between these two normally (Benedict and Cathcart,[17] Boothby[18] and Lindhard[19]). In figure 3, the extra

17. Benedict, F. G., and Cathcart, E. P.: Carnegie Inst. Washington, pub. 187, 1913.

18. Boothby, W. M.: Am. J. Physiol. **37**:383, 1915.

19. Lindhard, J.: J. Physiol. **57**:17, 1922.

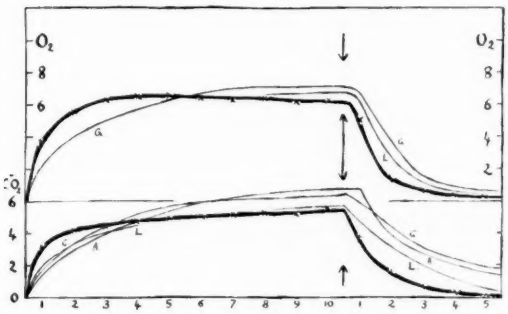

Fig. 1.—The oxygen intake and carbon dioxide output during ten minutes of light exercise and five minutes after. Extra oxygen and carbon dioxide are shown as cubic centimeters per kilogram of body weight to allow comparison between the four subjects. The upper curves (with the right hand scale) show extra oxygen intake; the lower (with the left hand scale) show extra carbon dioxide output. The thick lines on which separate points are shown as crosses are the curves for a normal subject and the thin are for three patients with heart disease. The arrows indicate the end of exercise. For clarity the curves during exercise are omitted when they are nearly normal, and A's oxygen curve is omitted entirely for the same reason. During the second minute after the exercise, the curves from above downward are those of G, L and a normal person for oxide and of G, A, L, and a normal person for carbon dioxide. The curves show relatively small changes as regards oxygen intake but some increase after the exercise, and a smaller output of carbon dioxide at the beginning and a much greater output after the exercise is finished.

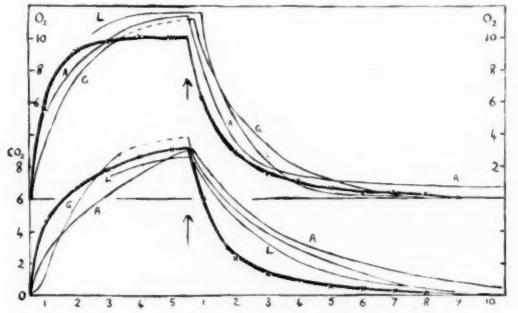

Fig. 2.—The oxygen intake and carbon dioxide output during five minutes of harder exercise and for ten minutes after. The scale is the same as in figure 1. During the second minute after exercise the curves from above downward are those of G, L, A and a normal person for oxygen and A, G, L and a normal person for carbon dioxide. At this harder exercise the patients' oxygen intake is greater than normal except just at first, and more is taken in after the exercise is finished. The curves for carbon dioxide show a much smaller output at the beginning except for L and a much larger output at the end of the exercise.

consumption of oxygen has been plotted against the work, expressed as kilogram meters per minute, for the normal subject when taking three, six, nine and twelve steps a minute, and for patients when taking six and twelve steps a minute. The curve showing the relationship was practically a straight line, and there was not much difference between the oxygen intake of the normal subject and that of the patients. Owing to the lighter weight of the patients, the work they did for the same rate of exercise was less, but the oxygen intake for the same work was a little more. The line was steeper, showing a faster increase of oxygen intake as the work increased, but the close general agreement meant that the diminished capacity of the patients for increasing their oxygen intake, rather than any change in the metabolism or oxygen requirement, caused breathlessness.

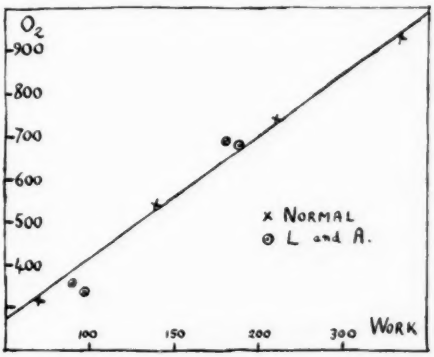

Fig. 3.—The extra oxygen intake (per minute) for increasing work (as kilogram meters per minute). Oxygen is shown as cubic centimeters per minute and the work done as in kilogram meters. The four points shown for the normal subject represent three, six, nine and twelve steps a minute. The two points shown for each of two patients (whose weight was about two thirds of the normal) represent six and twelve steps a minute. The close agreement between the straight line joining the four points for the normal person and those joining the points for the patients with heart failure shows that it is not any change in the metabolism or the oxygen requirement which is responsible for their breathlessness, but simply their diminished capacity for increasing their oxygen intake.

THE OUTPUT OF CARBON DIOXIDE DURING AND AFTER EXERCISE

All that has been said about the oxygen intake applies with greater force to the output of carbon dioxide. At both rates the amount of carbon dioxide expired relative to the work was always greater in the patients with heart disease than in the normal subject, and, except in one at the slower rate, the difference was always considerable. For the slower rate there was little difference during, but much more after, exercise, the normal subject getting rid of less than 13 per cent of the total after the exercise and the others of 23, 30 and 28 per cent,

respectively; for the faster rate the figures were 25 for the normal subject and 38, 45 and 51 for the patients.

If the results are followed from minute to minute (figs. 1 and 2), the output of carbon dioxide was more consistent than the oxygen intake. The main difference was the smaller output in all three patients during the first two or three minutes of exercise and the greater output after stopping, sometimes for five minutes, sometimes longer, the increase at the end being much more than the decrease at the beginning. In the normal subject the output reached almost its maximum by the third minute of exercise but continued rising slightly, while after stopping it reached nearly its resting value by the third or fourth minute. At the slower rate the changes were most striking in A. and G., L. being nearly normal by the fourth minute of exercise and by the fourth minute after stopping. The output was higher in A. and G. even during the latter part of the exercise. At the faster rate the difference at the beginning was greater, especially in A. and G., while after exercise it was greater and lasted longer, being considerable in all three patients five minutes after the exercise was stopped.

The output of carbon dioxide depends on two variables: the total ventilation and the percentage of carbon dioxide in the expired air. At rest the latter was much lower in G. and A. than in a normal person. The greater output required by the harder exercise was obtained mainly by increasing the ventilation so that there was little difference in the percentage of carbon dioxide. The first minute after the exercise was finished, the percentage was often a little higher, but it then fell to near the resting value at about the same rate as the ventilation.

The behavior of L. was most like the normal, as her percentage at rest and her rise during exercise were only a little low and well within normal limits (4 instead of 4.3). In G., whose reaction was most abnormal, the percentage fell steadily throughout the exercise and, as might be expected from this, was lower with the harder than with the easier exercise, namely, 2.2 and 2.4 from 2.55 at rest. This meant that his ventilation had to be increased more than in the others to get an adequate output of carbon dioxide, as was found at both rates of exercise. He could not continue the harder exercise for more than three minutes, and his shallow rapid breathing with such a low percentage of carbon dioxide in the expired air was no doubt closely associated with his greater disability. A. occupied an intermediate position. The percentage was as low as in G. at rest and was still low during exercise, but the increase was about the same as in health. As his breathing was deeper than normal, this was not even a partial reason for the low percentage of carbon dioxide.

PULMONARY VENTILATION DURING AND AFTER EXERCISE

Little need be said about this, as it has been discussed in a previous paper.[15] The rise and fall of the curves for the ventilation are in general similar to those shown in figures 1 and 2 for the carbon dioxide output. The rate and depth of breathing are given in table 3. In L. the breathing was slightly, and in G. considerably, more rapid and shallow; while in A. it was strikingly deeper and slower, as has been found frequently but not invariably in patients with aortic regurgitation. The probable explanation of the shallow breathing so often occurring with

TABLE 3.—*The Rate and Depth of Breathing During and After Exercise*

Time		Exercise: Six Steps a Minute (10 Minutes)								Exercise: Twelve Steps a Minute (5 Minutes)							
		Normal		L.		G.		A.		Normal		L.		G.		A.	
	Min.	Rate	Depth	Rate	Depth	Rate	Depth	Rate	Depth	Rate	Depth	Rate	Depth	Rate	Depth	Rate	Depth
Rest.....	..	16	430	19	310	23	220	14	585	16	425	17	355	23	220	14	585
During exercise...	1	18	685	19	435	25	240	15	565	18	735	23	455	30	240	13	880
	2	19	730	20	480	31	260	16	610	21	775	22	585	45	265	13	1050
	3	19	725	20	480	36	255	15	750	21	855	21	645	44	330	15	1020
	4	19	735	20	505	37	280	16	800	23	855	22	640	..	...	16	1100
	5	19	770	20	505	36	295	14	915	23	910	22	670	..	...	18	1060
	6	20	725	21	495	37	310	16	865	..	...	..	...	..	...	..	....
	7	20	735	21	510	38	305	16	900	..	...	..	...	..	...	..	....
	8	20	760	20	535	38	315	16	900	..	...	..	...	..	...	..	....
	9	20	745	20	520	39	320	16	930	..	...	..	...	..	...	..	....
	10	20	760	20	545	41	315	16	980	..	...	..	...	..	...	..	....
Rest after exercise...	1	18	670	17	580	32	310	16	920	18	900	19	655	37	345	17	1030
	2	18	530	17	550	31	310	15	750	17	625	18	620	35	300	15	990
	3	18	455	17	470	30	280	13	775	17	525	18	540	33	290	15	835
	4	17	440	19	405	28	255	13	715	17	465	17	500	31	260	15	770
	5	17	440	19	350	26	270	14	685	16	450	18	430	31	205	15	750
	6	16	455	19	315	27	250	13	700	16	435	17	405	31	205	15	725
	7	16	430	20	305	26	240	14	610	16	435	17	375	30	190	14	720
	8	16	455	20	300	26	215	14	590	16	430	19	340	28	170	14	685
	9	15	445	20	295	25	215	14	570	16	430	18	330	26	185	14	740
	10	15	440	20	295	25	210	14	570	16	430	18	355	26	185	14	650

mitral stenosis is in the greater liability to pulmonary congestion, and in several patients one of us has found that the shallow breathing is associated with a greatly reduced vital capacity.[15] Other work on this subject has been discussed in this paper and further observations are being made. The importance of vital capacity in the breathlessness of heart disease has been emphasized by Wentworth and Peabody,[20] Hewlett,[21] Myers [22] and others.

20. Peabody, F. W., and Wentworth, J. A.: Clinical Studies on Respiration, Arch. Int. Med. **20**:443 (Sept.) 1917.

21. Hewlett, A. W.: Heart **11**:195, 1924.

22. Myers, J. A.: Vital Capacity of the Lungs, Baltimore, Williams & Wilkins, 1925, p. 39.

The drum records of the ventilation were characteristic for each patient, and three typical tracings are shown in figure 4. The ventilation of A. rose from 8.5 liters at rest to 17.2 liters during the third minute of exercise, and that of G. from 5.1 to 14.4. The rate in A. rose only from 14 to 17.5, while in G. it was already fast at rest and rose from 25 to 43. There was an equally striking difference in the depth of breathing, and these two results were characteristic of what has usually been found in aortic regurgitation and in mitral stenosis, respectively. L., whose disability from mitral stenosis was less severe, occupied an intermediate position. The shallow, rapid breathing of G. and the slow, deep breathing of A. are well shown in these tracings, as is the sudden increase at the beginning and the slow decrease at the end of the exercise. It is clear that in these patients (as Krogh and

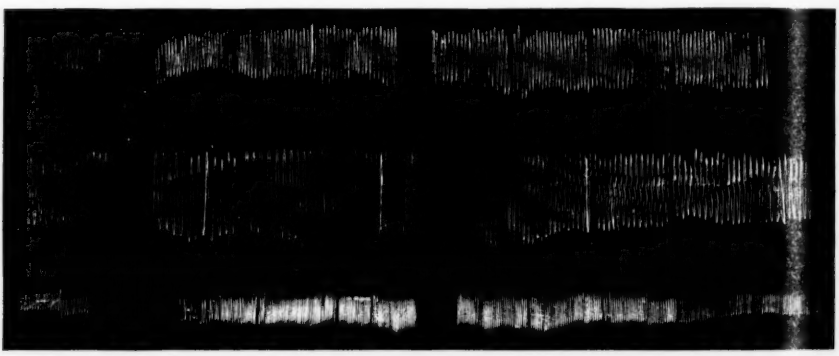

Fig. 4.—Respiratory tracings before, during and after exercise (twelve steps a minute for five minutes) in three patients with heart disease. The record has been cut to show the results of one minute at rest before exercise, the first three minutes of exercise and five minutes of rest after the exercise. The drum was running nearly, but not quite, at the same rate in each subject. The top record is from L with mitral stenosis, the middle from A, whose main lesion was aortic regurgitation, and the bottom from G, with mitral stenosis. The writing point was raised at the end of each minute. The figures for the ventilation and the rate and depth of breathing are given in tables 1, 2 and 3.

Lindhard [1] and others have shown in normal subjects) the increased breathing, practically coincident with the onset of exercise, must have been brought about by the nervous system, while the waning at the end controlled by the more usual chemical stimulus.

RESPIRATORY QUOTIENT DURING AND AFTER EXERCISE

Little change was noted in the respiratory quotient of the normal subject with the exception of a small drop in the second minute of exercise and a corresponding rise toward the end of the harder exercise and in the second minute after stopping the easier one. In patients

at rest the quotient was within normal limits, but at the beginning of exercise it dropped below 0.7 in each case and remained below the normal, except that in A., when he continued to exercise for ten minutes, it rose above 0.9 toward the end, as it did in the normal subject during harder exercise. After stopping, the changes were greater, and in all the patients the rise was higher and lasted longer. This was especially well shown with the harder exercise and with A. at both rates, the quotient being above 1 for about five minutes. These results are shown graphically in figure 5. Presumably owing to the defective circulation, the carbon dioxide produced cannot be eliminated at the beginning, and the retention leads to increasing breathlessness and to a

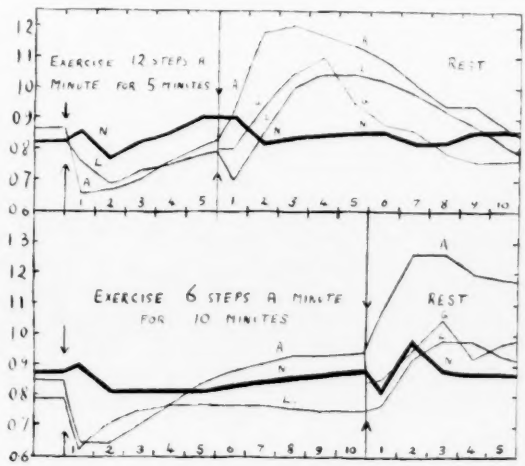

Fig. 5.—Respiratory quotient during and after exercise. The thick lines are for the normal person, and the thin lines for three patients with heart disease. (For clarity one of these is shown only after the exercise). The arrows indicate the beginning and end of exercise. In the normal subject there was no great change. In all the patients at the harder, and in A, at both exercises, there was a drop at the beginning of and a considerable rise after the exercise.

slow fall in the ventilation with consequent washing out of carbon dioxide after the exercise is finished.

Taking the total metabolism for the easier exercise and for ten minutes after, the quotient for the normal subject was 0.84; for L., 0.79, and for G., 0.82—figures not far removed from the values at rest—but for A. the figure was considerably higher, namely, 0.92. The corresponding figures for the harder exercise were: for the normal subject, 0.83; for L., 0.83; for G., 0.8, and for A., considerably higher, 0.9. If the quotients for the extra metabolism are considered after deducting the values at rest, they were hardly different, except for G., in whom they were raised to 0.95 and 0.98, respectively.

SUMMARY AND CONCLUSIONS

A detailed investigation of the effect of exercise—roughly equivalent to slow and ordinary walking—has been made in three patients with heart disease. The metabolism relative to the subject's weight did not vary greatly from the normal, which shows that the capacity for increasing ventilation and circulation, rather than any change in the metabolism or total ventilation required, limits exertion. Throughout the range examined, both of these increased regularly with the work done, so that their relationship could be shown graphically by a straight line.

In these patients the extra oxygen intake as the result of work was only slightly higher than normal. As the exercise became harder the percentage of the extra oxygen required, which had to be obtained at the end of the exertion, increased more in the patients. Both these differences would tend to produce breathlessness, but the degree of change would not have been sufficient to do so in a normal subject.

Greater differences were noted in the elimination of carbon dioxide. At the end of exercise it was retained more than normally, so that the output of this gas and the ventilation fell to their resting values more slowly. The changes at the beginning were similar but less striking. Consequently, the respiratory quotient fell rather more than usual at the beginning and rose considerably more than usual at the end. The percentage of carbon dioxide in the expired air tended to be low, probably because over-ventilation was needed to compensate for defective circulation. In one patient there was practically no rise in the percentage during exercise, so that the whole of the increased output of carbon dioxide had to be obtained by increased ventilation.

The rate and depth of breathing varied in type. In a patient with mitral stenosis it was rapid and shallow. This has been found in several others with mitral stenosis, and is probably associated with the reduced vital capacity and congestion of the lungs to which these patients are liable. The rapid, shallow breathing is itself a further cause of breathlessness. In a patient with aortic disease the breathing was deep and of normal rate. This has been found frequently but not invariably in patients with aortic regurgitation.

Book Reviews

DIE ZUCKERKRANKHEIT UND IHRE BEHANDLUNG. By C. VON NOORDEN, Geh. Medizinalrat, Professor at the University, Frankfurt, a. M., and I. Issac, Professor at the University, Frankfurt, a. M. Eighth edition. Cloth. Price, $12.40. Pp. 627, with 30 illustrations. Berlin: Julius Springer, 1927.

In the ten years that have passed since the appearance of the seventh edition of von Noorden's classic work, revolutionary developments have occurred in the field of diabetes, and these have necessitated a complete revision as well as considerable amplification of the text. Much of this development has originated in countries rarely heard from on the subject of diabetes before 1917. The field of activity in the research and therapy of diabetes has been extended to such a degree since 1917 that the German clinicians no longer so completely monopolize the subject. It is doubtful, therefore, whether this eighth edition of "Die Zuckerkrankheit" will have as great an influence on American readers as previous editions.

The survey of the literature is highly praiseworthy and, although the chapters on dietary management may be prized by many chiefly for their historical interest, the parts of the work bearing on symptomatology and complications must continue to command the greatest admiration. A section on the diseases of the eye in diabetes has been contributed by E. Grafe. This is based on examinations of the eyes in 1,200 cases and represents the most comprehensive study of this subject that has appeared.

The chapters on the theory of carbohydrate metabolism deserve careful examination by American students of this subject, particularly because thought in this country has been so largely molded by the school which has taught that the chief fault in diabetes was defective oxidation of sugar. Von Noorden has maintained, since 1910, that this teaching is wrong; that the tissues in diabetes oxidize sugar normally, and that the difficulty lies in an oversensitiveness of the liver whereby abnormal amounts of protein and fatty acid are converted into glucose (overproduction of sugar; sugar from fat). The action of the pancreas (insulin) is to check the activity of the liver, inhibit the formation of sugar from protein and fat and stabilize the glycogen stores of liver and other tissues. This theory, resting until recently on unsatisfactory supports, has been considerably strengthened by the developments of the last few years. Thus Frank Mann's method of resecting the liver, in a two-stage operation which permits the animal to recover from the shock of the operation and makes it possible to obtain dependable respiratory quotients after the resection, has provided evidence that the quotients tend to rise toward unity after hepatectomy. The results of his experiments are much more reliable than were those of the earlier experiments of Porges and Salomon. This was the original experimental basis of von Noorden's theory, Porges' and Salomon's results being taken to mean that, in the absence of the liver, fat and protein cease to be sources of energy.

It has also been demonstrated fairly conclusively in late years that muscular work is accomplished only with carbohydrate. This should be credited to A. V. Hill and Meyerhoff's Nobel prize contributions; curiously, A. V. Hill's name fails to appear either in the text or in the bibliography. If Hill and Meyerhoff's conclusions stand, sugar must be supplied to the muscles from fat whenever the organism is subsisting on materials poor in carbohydrate. Recent work of Embden, Lesser and others seems to establish the fact that surviving diabetic tissues in perfusion experiments utilize glucose as well as do the tissues of normal animals; finally, the phenomenon of insulin shock

is readily explained on the overproduction theory, but causes serious embarrassment for those who hold to the view that oxidation is deficient.

In discussing the etiology of diabetes, von Noorden and Isaac place great emphasis on heredity, not only heredity for diabetes alone but also heredity of inferiority of the endocrine system as a whole. One member of the system may be affected in one person of a family and another member in the next. Hereditary diabetes in children is not found to be milder than other juvenile diabetes, contrary to the views of Naunyn Loeb and Joslin. Inbreeding, restriction of the field of natural selection, is thought to be the explanation of the high incidence of diabetes in the Jews and high-cast Hindus. An endogenous disposition (abiotrophy) is the underlying cause of diabetes, while exogenous factors merely play the rôle of unleashing (auslösende) agents. Infectious diseases rank first among these. That such exogenous factors, infections, overeating and nervous shocks can cause diabetes in the absence of biologic insufficiency, is considered doubtful.

SULFUR METABOLISM. By MAX KAHN and FREDERIC G. GOODRIDGE. Price $9. Pp. 800. Philadelphia: Lea & Febiger, 1926.

This work is a compilation of references on sulphur metabolism rather than a review, and this is essentially all that the authors claim for their publication. This volume is comprehensive in scope. The first five chapters are devoted to tables and statements concerning the sulphur content of foods, excreta, secretions, tissues and pigments. The next seven chapters are devoted to data on sulphur metabolism in different animals as affected by age, various physiologic states, drugs and diseases. The following ten chapters are devoted to more detailed discussions and citations on various specific forms of sulphur, such as protein sulphur, lipin sulphur, glucosides, ethereal sulphates and other forms. Brief chapters on hydrogen sulphide as related to biology, on mercaptans, on sulphur bacteria, sulphur in soil and various synthetic sulphur derivatives of possible biologic interest cover the last 200 pages. The concluding chapter covers a brief summary on the methods used for determining total and various forms of sulphur in biologic material. Naturally the discussions are of typical encyclopedic character, and in no sense is the work critical.

TECHNIQUE DE LA REACTION DU BENJOIN COLLOIDAL. Par. GEORGES GUILLAIN, professeur a la faculte de medecine de Paris, membre de L'Academie de Medecine, medecin de La Salpetriere et Guy Laroche, medecin des Hospitaux de Paris; P. Lechelle, medecin des Hopitaux de Paris. Paris: Masson & Cie.

This is a small monograph of thirty-four pages, liberally illustrated with curves and four photographic reproductions of reactions. It is simply a report of the technical experiences of the authors covering a large series of observations; the theory and interpretations of the tests are not covered.

The test is based on principles similar to those on which the Lange gold reaction is based, and gives similar results. A much larger series of dilutions is used, generally sixteen, and the resulting curves are therefore more complicated and give promise of being of greater clinical value in the more unusual forms of cerebrospinal syphilis. The tabetic and paretic curves closely resemble the Lange curves.

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